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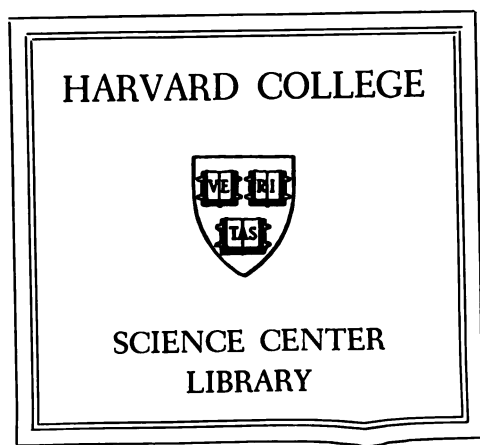
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BIOLOGICAL BULLETIN

EDITED BY

*THE DIRECTOR
AND MEMBERS OF THE STAFF*

OF

The Marine Biological Laboratory

WOODS HOLL, MASS.

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BIOLOGICAL BULLETIN.

SOME RELATIONS BETWEEN NERVOUS TISSUE AND GLANDULAR TISSUE IN THE TUNICATA.

MAYNARD M. METCALF.

It is well known that in the ascidians the definitive brain and the neural gland are both derived from the same region — the trunk region of the central nerve tube of the tadpole. The ganglion is derived from one wall of this tube, and the gland is derived from the opposite wall. Six years ago I pointed out that the ganglion of *Salpa* is homologous with both the ganglion and the neural gland of ascidians; the dorsal part of the *Salpa* ganglion corresponding to the ascidian brain, and its ventral part corresponding to the ascidian neural gland.¹ That is, a certain portion of the embryonic nervous system in ascidians becomes transformed into the neural gland, while in *Salpa* the corresponding region does not suffer this change, but remains as part of the definitive brain, its cells functioning as gland cells in the adult. I have recently found in the ascidians an interesting series of diverse conditions as to the origin of the gangliated nerve which runs down in the median line of the partition between pharynx and cloaca.

In this region, the dorsal raphe, one finds a large blood sinus, a muscle (either single or double), a gangliated nerve cord (the rapheal nerve), and frequently a prolongation from the neural

¹ "The Eyes and Sub-Neural Gland of *Salpa*," *Memoirs from the Biological Laboratory of the Johns Hopkins University*. Vol. ii, Part iv. 1893.

gland, which I have called the rapheal duct. In different species the rapheal nerve may arise from the cellular cortex of the brain, from the neural gland, or from a mass of cells formed by the fusion of the brain and the gland. The five accompanying diagrams show some of the conditions found.

In *Cynthia papillosa*, Fig. 1, the rapheal nerve arises from the cellular cortex of the brain. Alongside it in the raphe is found the unusually large rapheal duct, which has extended down from near the posterior end of the epineural gland. The rapheal duct and rapheal nerve are wholly distinct.

In *Distaplia magnilarva*, Fig. 2, there is no rapheal duct. The brain and neural gland are united posteriorly. The rapheal nerve arises from the cortex of the brain, a little behind the point of fusion of the brain with the gland.

In *Amaroeicum constellatum*, Fig. 3, we find a rudimentary rapheal duct starting back from the gland, but it loses its lumen before going far, and then its cells become united with the cells of the brain to form a common mass of cells whose origin, whether from the brain or the gland, we are unable to determine. From this common mass of cells the *ganglion cells* of the rapheal nerve are derived, its *fibers* coming from the right posterior siphonal nerve.

In *Ascidia atra*, Fig. 4, we have a similar origin for the fibers of the rapheal nerve, but find an interesting difference in the derivation of its ganglion cells. A cord of cells pushes out from the dorsal surface of the brain, near its posterior end, and, after running back a short distance, unites with a backward prolongation of the gland, which runs up to meet it. The prolongation of the gland is evidently the rapheal duct. The two cords fuse immediately, the duct losing its lumen. The single cord of cells thus formed runs back some distance and then bends down to accompany the fibers of the rapheal nerve. Its cells soon become loosely arranged among these nerve fibers and are clearly the ganglion cells of the rapheal nerve.

In *Phallusia mammillata*, Fig. 5, these organs are exactly similar, except that the prolongation from the dorsal surface of the ganglion does not unite with the rapheal duct, but bends forward, soon ending blindly. In this species, then, the ganglion

cells of the rapheal nerve are derived solely from the rapheal duct, which is a prolongation from the neural gland.

The ganglion cells of the rapheal nerve in *Phallusia* have therefore had a roundabout history. Certain cells of the larval nerve tube were pushed out to form the neural gland. A portion of these gland cells extended backward until they came in contact with the fibers of the rapheal nerve.¹ Here they lose their regular arrangement and become the ganglion cells of the nerve. There is no evidence that these particular cells, even though a part of the gland, were ever functional as glandular cells. The corresponding cells, however, in many other species are functional gland cells. (Compare *Cynthia papillosa* above.)

The facts referred to in this paper show a peculiarly intimate relation between glandular tissue and nervous tissue in the *Tunicata*, hardly to be paralleled elsewhere in the animal kingdom.

THE MARINE BIOLOGICAL LABORATORY,
WOODS HOLL, MASS.,
July 20, 1899.

¹ This is based upon the assumption that the rapheal duct arises as a down-growth from the definitive gland rather than by the metamorphosis *in situ* of the trunk portion of the nerve tube of the tadpole. The rapheal nerve in *Salpa* and probably in ascidians arises as a down-growth from the brain. It is probable that the rapheal duct arises in a similar way as a down-growth from the definitive gland.

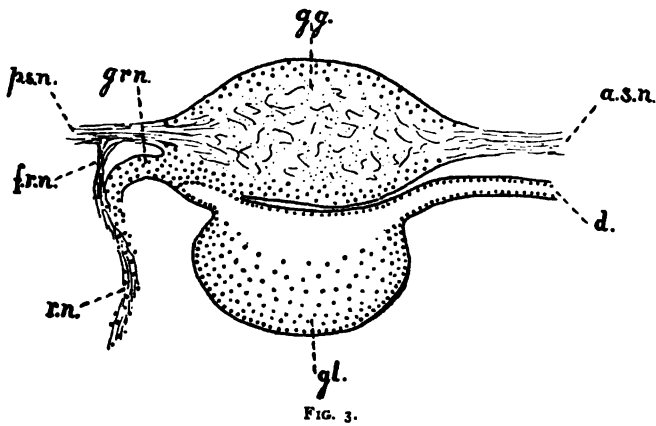
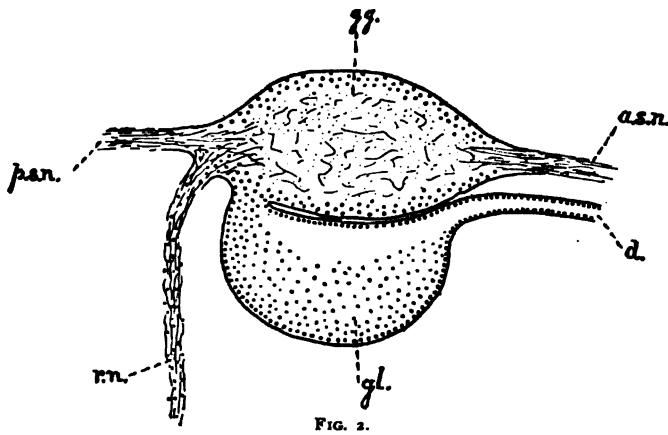
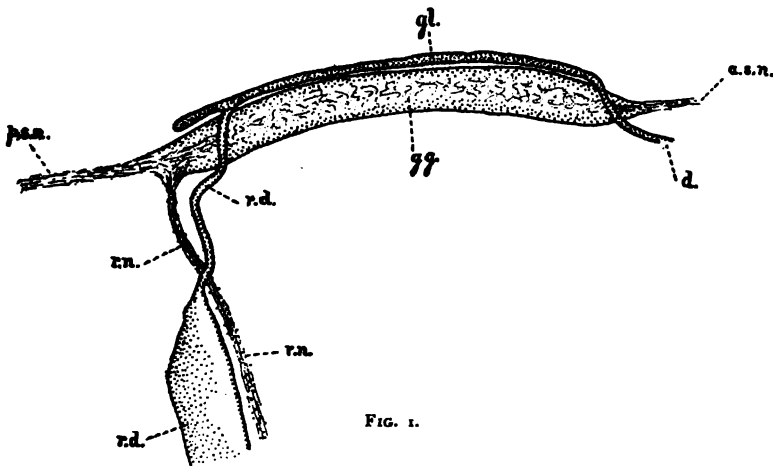
EXPLANATION OF FIGURES.

Reference Letters.

- a.s.n.* = anterior siphonal nerve.
d. = duct from neural gland to ciliated funnel.
f.r.n. = fibers of rapheal nerve.
g.c. = cord of ganglion cells.
gg. = ganglion (brain).
gl. = neural gland.
g.r.n. = ganglion cells of rapheal nerve.
p.s.n. = posterior siphonal nerve.
r.d. = rapheal duct.
r.n. = rapheal nerve.

The figures are diagrammatic parasagittal sections of the ganglion and neural gland, a little to the right of the median plane. In Figs. 4 and 5 the rapheal duct, which in reality lies on the right surface of the ganglion, is shown, although in reality it lies much to the right of the plane of the rest of the section.

- FIG. 1. *Cynthia papillosa*.
FIG. 2. *Distaplia magnilarva*.
FIG. 3. *Amaroecium constellatum*.
FIG. 4. *Ascidia atra*.
FIG. 5. *Phallusia mammillata*.



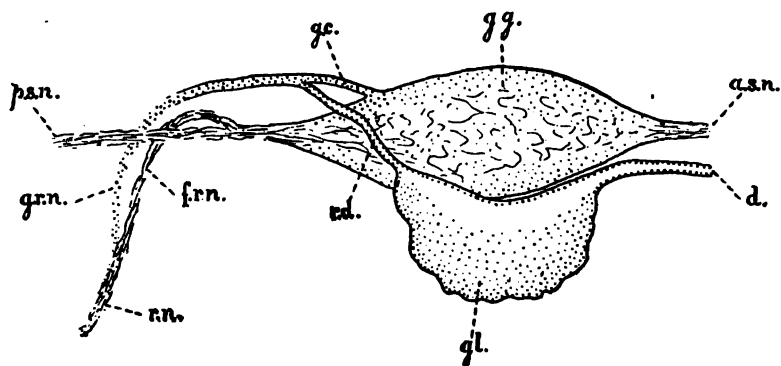


FIG. 4.

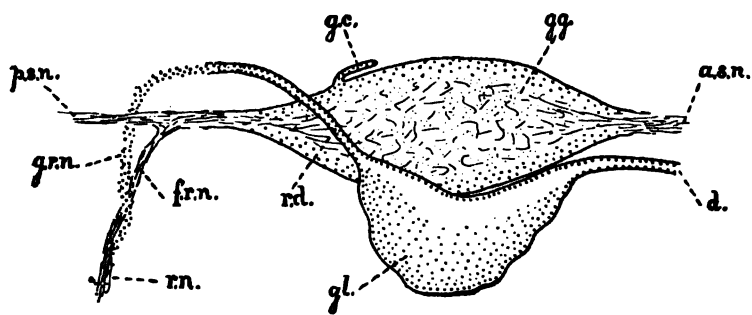


FIG. 5.

REGENERATION OF TISSUE COMPOSED OF PARTS OF TWO SPECIES.

T. H. MORGAN.

BORN's experiments in grafting together tadpoles of different species of frogs have demonstrated that each part, whether large or small, retains the characteristics of the species to which it belongs. During the development of an animal, formed by the union of parts of two species, the tissues do not influence each other, but each develops its own specific peculiarities.

Joest has shown that when parts of two different species of earthworms are grafted together each part retains its specific characters. He has further shown that if, after grafting, a portion of one of the parts is cut off, the new part that is regenerated is like the part from which it immediately arises, and is not influenced by the part belonging to the other species, even when the latter is very large, and the former (that from which the new part arises) is very small.

Many experiments have been made with plants in which different species have been grafted together, and the subsequent growth of the two parts studied. Vöchting, who has given a detailed account of these experiments and has made others himself, has shown that in general no influence of a specific character is transmitted from one part to the other, although in certain cases¹ the parts do have some influence on each other.

The following experiments were made, not so much to determine whether the tissues of one component of a graft influence the kind of regeneration of the other, since this point seemed fairly settled by Joest and by Harrison, but I hoped to find out if new tissue, made up of cells derived from parts of two species, showed any mixing of the specific characters of the two species.

¹ Particularly in those cases where annual and biennial varieties are grafted together.

It seemed possible, at least, that new tissue, composed of cells derived from two species, might show the influence of its dual origin.

Harrison¹ has shown that the tails of young tadpoles may be interchanged even when two species are used, and that later the ectoderm of the body of the larger component grows out over the base of the grafted tail, slipping over the region where the tail has been grafted on, as shown in Fig. 1. If two species be used, and then, after the tail has grown to the stage shown in this figure, the tail be cut off just distal to the point of union, as shown in Fig. 2 by the vertical line, there will be present at the exposed end two kinds of tissue — the ectoderm, which is the same as that covering the body of the tadpole, and the inner tissue, composed of muscles, connective tissue, pig-

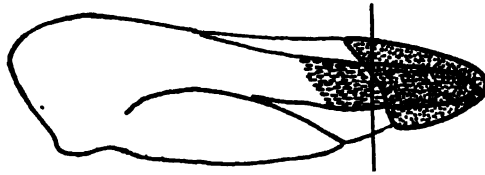


FIG. 1. (After Harrison.)

ment cells, notochord and nerve cord, that belong to the grafted tail. Under these circumstances the new tail that regenerates will be made up of parts of two species. Harrison carried out an experiment of this sort. He writes² in regard to the result: "The tail of a larva of *R. virescens* was replaced by the tail of a larva of *R. palustris*, in the manner described above. Forty-eight hours later, at which time the sketch was made (see Fig. 1), the tail was amputated. The epidermis from the *virescens* body had then pushed out considerably over the root of the tail, so that in cutting, almost all of the grafted epidermis (stippled in the figure) was removed. But a considerable portion of the underlying organs of the transplanted piece (shaded in the figure) remained, and it was from this component that regeneration took place in all the tissues, with the exception of part of the epidermis. The newly grown tail was

¹ Harrison, R. G., "The Growth and Regeneration of the Tail of the Frog Larva," *Roux's Archiv.* Vol. vii, 1898.

² Page 473. Case 13.

of normal form, and, as far as could be observed, it had the characteristics of the species of the grafted stump (*palustris*) and not those of the body (*virescens*). This was seen in the character of the pigmentation, and especially in the absence of the large black blotches along the side of the tail, which are found constantly in the regenerated appendages of *R. virescens*. In spite of the insignificant size of the grafted stump, as compared with the whole body, and in spite of the fact that the nourishment conveyed to the growing appendage is brought there in blood, which is largely derived from the body, the tissues maintain their specific characters.¹"

By using two species in which there is a marked difference in the pigmentation of the ectoderm and also some distinctive difference in the color of the pigment cells in the mesoderm, I hoped to be able to determine more definitely the character of the new part, and further, by observing the tissues of the two species, where they are regenerating side by side, to see if they mutually influence each other. The problem is somewhat different from the one Harrison examined, since I was less concerned with the influence of the major component on the new regenerating part than with the possibility of a mutual influence of the new cells on each other. Harrison has shown, with some degree of probability, that the former influence is not shown in the new part, but the latter problem is not specially considered. X

I have found it possible to graft together two such differently pigmented tadpoles as *Rana (temporaria) sylvatica* and *R. palustris*. The former breeds earlier, but the development will be retarded several weeks if the dishes in which the tadpoles are placed be put on ice in an ice chest. It is better to let the tadpoles develop as far as the stage when the tail-knob is just about to appear, since at this stage they withstand better

¹ "I had hoped to obtain more definite evidence concerning the influences which regulate regeneration, from experiments carried out along these lines. But, owing to unfortunate circumstances, most of the larvae of this series died. Besides, all regenerated tails deviate somewhat from the normal type, especially as regards pigmentation, which fact would bring in a considerable element of uncertainty, and in the tail I have not been able to find any other characters which could with safety be considered diagnostic of either species."

the effect of the ice-cold water. If segmenting eggs or the early gastrula stages be put on ice they are killed after several days, although the latter stages withstand the cold longer than the former. The young tadpoles do not seem to be in the least injured, and may even slowly continue to develop, but at so slow a rate that after three weeks the tail had grown only a very little. Since in this locality the eggs of *R. palustris* can be obtained in great abundance for a period of at least two weeks, I have had plenty of material of both species.

The tadpoles were operated upon at the time when they had reached the age shown in Harrison's Fig. 2. They were still in the jelly membranes. Muscular movements of the body had scarcely begun at this time. The tadpoles were held in place by small pieces of aluminium wire. Silver wire used by Born and by Harrison would probably be better, since it is heavier.

The young tadpole of *R. sylvatica* is very black, the color being due to the deeply pigmented ectoderm and to some extent to pigment in the mesoderm. The young tadpole of *R. palustris* is much lighter in color. The ectoderm contains a yellowish pigment, and the pigment cells of the mesoderm are lighter in color than those of the other species. After grafting together parts of these two species, the difference in color of the two parts is so marked that it can be easily seen with the naked eye. Under the microscope one can tell readily whether an individual cell in the ectoderm belongs to the one or to the other species. In later stages, when the ectoderm has become clearer, the two kinds of cells can no longer be distinguished without a microscope. The core of the tail of *R. sylvatica* is much darker than that of *R. palustris*, and the line of union of the two can be seen with the unaided eye.

As the tadpole grows larger, it will be found that the ectoderm of the smaller component grows less rapidly than the rest of the tail, and as a result the ectoderm of the larger component extends over the base of the grafted tail, as Harrison has stated (Figs. 2 and 3). The tadpoles were allowed to grow for about ten days, or somewhat longer,¹ and then the tail was cut off in various ways.

¹ It would have been better to have cut the tail off sooner, since the difference in the ectoderm of the two species is less marked in later stages.

Experiment I.—The tail of *R. palustris* had been grafted upon the body of *R. sylvatica*. The tadpole appeared at the time of the second operation, as shown in Fig. 2 A (April 25). The dark ectoderm of the major component—*R. sylvatica*—had grown out over the base of the tail of the smaller com-

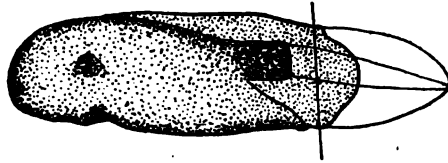


FIG. 2 A.

ponent (*R. palustris*). The region of union of the inner tissue can be seen where the dark and the light parts meet. The tail was then cut off, as shown by the vertical line in the figure. In consequence, there was left exposed at the cut end of the tail

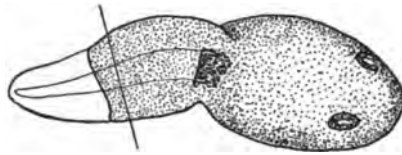


FIG. 2 B.

the inner tissues derived from *R. palustris*, and the outer from *R. sylvatica*. A new tail began to regenerate, and during all of its subsequent development the new tail was made up of ectoderm exactly like that of the major component, and of inner

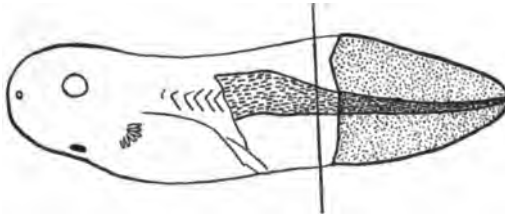


FIG. 3.

tissue whose pigment cells resembled those of the minor component. In other words, both inner and outer tissues regenerated their kind and showed no commingling of characters.

Experiment II.—In this experiment the major component was *R. palustris* and the minor *R. sylvatica*. After the new

tail had reached the stage shown in Fig. 3, it was cut off as indicated by the vertical line. There was left exposed at the cut end the light-colored ectoderm of the major component and the inner tissues of the minor component. The new tail that developed had light ectoderm on the surface and a dark interior. Each part regenerated its specific tissue and was uninfluenced by the developing tissue of the other species.

Two points present themselves for consideration. If the tail of an ordinary tadpole be cut off and subsequently develop, does the regenerated tail show the specific characters of the normal tail or is it different? I have examined the regenerated tails of both species and find that both the ectoderm and the mesodermal pigment cells are like those of a normal tail. It is, however, not very uncommon, both in regenerated tails of normal tadpoles and also in grafted tadpoles, to find the mesodermal pigment cells imperfectly developed, and in such cases the specific character of the cells is not obvious; but in all cases in which the pigment cells are well developed, the specific character is readily seen, especially in the cells lying along the central part of the tail. It should be stated, however, that I have occasionally found isolated cells whose character was doubtful, but the large majority of cells are unquestionably like those of the tissue from which the new tail arises. The second question is whether the ectoderm forms new cells over the new part, or does the old ectoderm simply extend out over the new part? There is the appearance in the regenerating tail of the formation of a new ectoderm at the tip of the new tail, where the cells are more crowded together and smaller than over the base of the tail. It is not improbable that in addition to this new ectoderm the old ectoderm extends also over a part of the new tail.

Experiment III. — In several cases the tail was cut off obliquely, in much the same way as in Harrison's experiment. Owing to the difference in pigmentation of the two kinds of ectoderm, I could follow the subsequent history of each and determine whether, along their line of contact, and in the region where new cells are developing, the specific characters of the cells are altered.

As shown in Fig. 4, the tail of a tadpole, in which the major component is *R. palustris* and the minor *R. sylvatica*, was cut off obliquely, leaving a small amount of the dark ectoderm of *R. sylvatica* on the upper side. The inner cells at the cut edge all belonged to *R. sylvatica*. When the new tail developed, it showed along its upper part the dark ectoderm of *R. sylvatica*, that had developed from the small piece left at the time of the

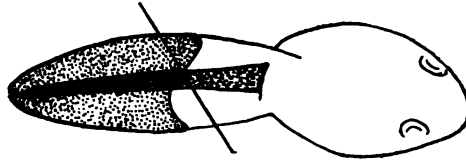


FIG. 4.

operation. The area covered by the dark ectoderm was greater than that left after the tail was cut off, but it cannot be stated how much of this increase is due to the cells becoming flatter and how much to new cells formed at the free edge.

In another similar experiment, in which, however, the major component was the dark species, *R. sylvatica*, and the minor the paler species, *R. palustris*, the tail was cut off (April 27), as shown in Fig. 5. A large area of light ectoderm was left on the dorsal surface of the tail, and only a small amount of the

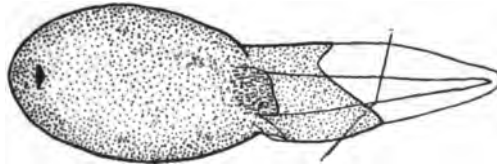


FIG. 5.

black ectoderm came to the edge of the lower part. On May 10, when the new tail was fairly well developed, it was found to have its upper surface covered by light-colored ectoderm, and its lower by dark ectoderm, while the interior mesodermal pigment cells were like those of *R. palustris*. Each tissue had regenerated its like, and the light ectoderm of the minor component showed no influence of the other, dark ectoderm, even along the line of contact where new cells were developing.

In addition to these experiments I have records of four others similar to Experiment I (Fig. 2, *A, B*); three others similar to Experiment II (Fig. 3); and four others in which the tail was cut off obliquely, leaving both kinds of ectoderm at the cut edge. In all cases the specific character of the new tissue was like that of the old tissue from which it arose.

At first the difference in the ectoderm of the two species is very marked, but as the tadpoles get older the ectoderm seems to flatten and become more transparent, so that in these tadpoles it is difficult to distinguish between the two kinds of ectoderm. But if the tadpoles are examined every day one can detect differences in the two kinds of ectoderm for a longer period than could be done by casual observations alone. Wherever the ectoderm has not spread out, particularly at the tip of the tail, the dark pigmented cells of *R. sylvatica* and the yellowish cells of *R. palustris* can be readily detected. The pigment cells in the mesoderm assume their characteristic arrangement during the older stages, and as the ectoderm becomes more transparent, the cells can be easily seen in the living tadpoles. The tadpoles were all kept under the same condition, so that the effect of light on the pigment cells would be approximately the same in all experiments.

Unfortunately the differences in pigmentation are the only specific characters that can be made out readily in these tadpoles, but I think there can be little doubt that if the cells retain their characteristic pigmentation they also retain their other peculiarities.

We may conclude with some degree of probability that during regeneration in a region where the cells have been derived from two different species, each kind of new cell retains the character of the cells from which it is derived, and the specific characters of the cells of one species are not transmitted to the cells of the other species, although the developing cells in the new tissue may be in actual contact.

DINOPHILUS GARDINERI (Sp. Nov.).

ANNE MOORE.

PRELIMINARY NOTE.

A NEW species of *Dinophilus* was found in the summer of 1897, by Dr. E. G. Gardiner, at Woods Holl. The pool in which it is found is an artificial one, 12 by 14 feet, dug about eight years ago to obtain the peat in the marsh. Sea water does not flow into it, but when the tide is unusually high it percolates in through the sand. The salinity of the water is therefore subject to great variation, for a run of low tides results in condensation through evaporation, while heavy rains dilute it. In May, 1898 and 1899, *Dinophilus* was found by Dr. Gardiner in abundance upon green algae floating on the surface of the water. In June, 1898, he kindly brought the animal to my notice and I began work upon it. At that time it was not abundant, and by July 16 had entirely disappeared. In 1899 only two specimens were seen after June 28. Both years the disappearance was coincident with a rainstorm, so that it is quite possible that the influx of fresh water may account for it. Other observers (Hallez,¹ Weldon,² Harmer³) have noted the periodical disappearance of *Dinophilus*. Weldon attributes it to the disintegration of the female, consequent upon the setting free of the ova, but Schimkewitsch⁴ maintains that eggs may be laid several times during the year, and that the female lives for some time after depositing them. He found special ducts present for carrying the eggs to the exterior, so that there

¹ Hallez, P., *Contributions à l'Histoire Naturelle des Turbellaires*. Lille, 1879, p. 155. (D. metameroides.)

² Weldon, W. F. R., "On *Dinophilus gigas*," *Quart. Journ. Micr. Sci.* Vol. xxvii. 1887.

³ Harmer, S. F., "Notes on the Anatomy of *Dinophilus*," p. 109, *Journ. Mar. Biol. Ass. of United Kingdoms*. New Series. Vol. ii, October, 1899.

⁴ Schimkewitsch, W., "Zur Kenntnis des Baues und der Entwicklung des *Dinophilus* vom Wei Ben Meere," *Zeit. für wiss. Zool.* Bd. lix. 1895.

is no necessity for disintegration to set them free. Of the disappearance of the animal, and a possible explanation of it, I will speak later.

This species of *Dinophilus*, which I take pleasure in calling *D. Gardineri*, differs in certain features from those species which have been noted by other observers. It is easily recognized without a lens, for its bright orange-red pigment makes a sharp contrast with the green algae upon which it is found. The average length of the form is about 1 mm., but under a

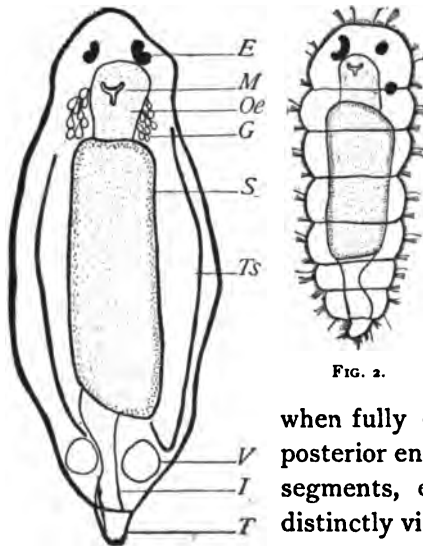


FIG. 1.

FIG. 2.

dissecting lens the highly colored intestine, with its characteristic stomach portion, and the red kidney-shaped eyes, are noticeable features. The body is about three times as long as it is broad, the proportions varying, of course, with extension or contraction; it is somewhat flattened dorso-ventrally, and when fully extended tapers toward the posterior end. It consists of six definite segments, exclusive of head and tail, distinctly visible in young individuals in a state of extension (Fig. 2). Neither young nor old individuals show segmentation when contracted, and old ones show it only when making a turn, not when moving rapidly in full extension. The head is rounded in front and bears the eyes on its dorsal surface (Fig. 1). The mouth is situated on the ventral surface posterior to the eyes at, or just anterior to, the union of the head with the first body segment. The small unsegmented tail approximates the length of a body segment. It is considerably narrower than the body and tapers to a point. The anus is situated dorsally to its base. Owing to the scarcity of material, I did not ascertain to my satisfaction the arrangement of the cilia, but as nearly as I could determine the animal is com-

pletely ciliated, and, in addition, each segment shows laterally two tufts of long cilia and a strong bristle anteriorly placed. These probably indicate the presence of two rings of cilia on each segment. The head bears two tufts of long cilia in front, and the tail bears several bristles. These are probably of a sensory nature.

No sexual dimorphism is present; it is impossible to distinguish the sex of young individuals. In mature females the paired ovaries are strongly colored and may be clearly seen.

This species differs from *D. gyrociliatus* (*apatris*) and *D. metameroides* in its lack of sexual dimorphism; in the number of segments and in the arrangement of cilia it differs from *D. gigas* (7 segments), *D. taeniatus* (5 segments), *D. pygmaeus* (5 segments), and *D. simplex* (4 segments); in the arrangement of cilia and in the possession of an unsegmented tail it differs from *D. vorticoides* (*caudatus*).

The bilobed or crescentic shape of the eyes of *Dinophilus* often looks as if they were on the way to becoming double, as is the case with some *Turbellaria*. I have found two specimens in which the right eye was made up of two spheres completely separated from each other. In one case they lay close together; in the other, one sphere was in the normal position, the other in the next segment (Fig. 2).

In explanation of the disappearance of *Dinophilus*, alluded to above, I have to offer an observation of a stage in its life history which to my knowledge has not been noted before. On June 27, 1899, in my search for specimens I came across capsules imbedded in the tangle of algae. Through the thin transparent walls I could distinctly see the characteristic form, color, and eyes of *Dinophilus*. In addition to the capsules, eight specimens were found on the same day. These were put by themselves in a shallow glass dish containing salt water and some algae, and were watered from day to day. At the end of a week only five specimens were seen; on searching for the other three, three capsules were found. I then realized that these capsules really represented an encysted stage of *Dinophilus*. The five remaining specimens were transferred to a fresh dish of clear water. Four of them disintegrated;

the fifth formed a cyst. Every stage of the process was watched. The animal became perfectly quiet, and a clear secretion was given off. After a time, probably from a sense of discomfort, it moved away from this secretion, leaving behind it an impression of its form. These impressions had been seen before, but it was not known to what they were due. After moving, the animal continued to give off the secretion, and at the end of the next day the capsule was completed, the whole process taking three days. The other individuals began to secrete in the same way, but in one case the animal was disturbed and the process stopped, and in the others they were attacked by protozoa, causing disintegration. The largest cyst that was found measured .5 mm., the smallest .13 mm. This decrease in size might easily account for their being overlooked, but in addition to this it was found that after keeping the cyst for a time the color faded out so that it became practically unrecognizable.

It is quite probable that this cyst is formed through the activity of numerous gland cells in the skin. Many observers have noted these glands, but no one has suggested an adequate function for them.

The affinities of *Dinophilus* and its systematic position make it a peculiarly interesting form, and I hope in a more favorable year to obtain sufficient material to complete my work upon it.

THE MARINE BIOLOGICAL LABORATORY,
WOODS HOLL, AUG. 6, 1899.

SOME MUSCINAE OF NORTH AMERICA.

GARRY DE N. HOUGH, M.D.

GIRSCHNER divides the *Muscidea* into two series—*Calypttratae* and *Acalypttratae*. The former he divides into two families, one of which is the *Anthomyidae*. Hypopleural bristles lacking. If three sternopleural bristles are present, they always have the arrangement 1 : 2 (*i.e.*, one in front and two behind). Ventral membrane usually present. Elbow of fourth longitudinal vein (if there is any) without an appendix.

This family Girschner divides into three groups :

1. *Coenosiinae*.—Fifth ventral segment of the male heart-shaped or split in the median line from the caudal border to a point beyond the middle. Fourth longitudinal vein straight. Abdomen usually elongate. Sternopleural bristles present. Squamulae separated from one another by an interspace that is broad to the very bottom ; squamula thoracalis never broadened towards the scutellum.

2. *Muscinae*.—Fifth ventral segment of the male with its caudal border straight or moderately concave (lunulate), at any rate not split beyond the middle, except in *Lispe*, where it is three-pronged. Fourth longitudinal vein straight or more or less bent up toward the third in the form of an apical cross-vein. Abdomen usually short or long oval. Sternopleural bristles present. Squamulae not separated from one another, in contact at their attached borders ; angle between them narrow and acute.

3. *Gastrophilinae*.—Sternopleural bristles absent. Fourth vein straight. Costal vein reaching only to or a little beyond the third vein. Squamulae but little developed, separated from one another by a projecting angle.

The *Muscinae* are divided into three sections :

a. *Muscinae coenosiaeformes*.—Front broad in both sexes.

Squamula thoracalis not broadened mesad and caudad. Wings not rilled.¹

b. *Muscinae ariciaeformes*. — Front narrow in the male, broad in the female. *Squamula thoracalis* not broadened mesad and caudad. Wings in the most recent forms (geologically speaking, the "youngest") rilled.

c. *Muscinae muscaeformes*. — Front as in b. *Squamula thoracalis* broadened out mesad and caudad as far as the edge of the scutellum. Wings rilled. Apical cross-vein present.

Girschner's family *Anthomyidae* includes, in the first two groups, the *Anthomyidae* and part of the *Muscidae* (sens. strict.) of other authors. The genera belonging to the former have been made the subject of a recent paper by Mr. Paul Stein in the *Berl. Ent. Zeit.*, Vol. XLII, pp. 151–288, 1897. This paper covers the *Coenosiinae*;² the *Muscinae coenosiiformes*; and the *Muscinae ariciaeformes*, except the genera *Myospila*, *Muscina*, *Clinopera*, *Hemichlora*, *Stomoxys*, and *Haematobia*. In these genera the fourth longitudinal vein is bent up, near its apical end, towards the third, and the arista is either pectinate or long plumose. They may be separated from one another as follows:

1. Proboscis long, slender, horny, adapted for piercing 2
 Proboscis not so constructed, provided at the tip with fleshy labellae 3
2. Palpi much shorter than the proboscis, arista pectinate *Stomoxys* Geoffroy
 Palpi nearly as long as the proboscis, arista pectinate, sometimes also
 with a few hairs below *Haematobia* Desvoidy
3. Arista pectinate *Hemichlora* v. d. Wulp
 Arista plumose 4
4. Sternopleural macrochaetae 2 : 2; eyes hairy *Myospila* Rondan
 Sternopleural macrochaetae 1 : 2; eyes not hairy 5
5. First longitudinal vein ends far beyond the middle of the costa. One
 or more well-developed pairs of anterior acrostichal bristles

Muscina Desvoidy

First longitudinal vein ends before the middle of the costa. No anterior
 acrostichal macrochaetae *Clinopera* v. d. Wulp

¹ These rills are very fine grooves in the surface of the wing which run in a sort of radiate manner toward the border. They are very numerous. A rilled wing denotes a higher stage of development, a more recent form, than an unrilled wing.

² Girschner's *Coenosiinae* includes a few genera which are commonly considered as members of the Acalyptrate family *Scatomyzidae*; these genera are not considered by Mr. Stein.

Stomoxys. — I have seen but one American species of this genus, which is the well-known "Stable Fly," *S. calcitrans* L. (Fig. 1, wing and chaetotaxy), very common both in Europe and this country. Of the species mentioned in Osten-Sacken's *Catalog*, *dira* Desv. and *inimica* Desv. are varieties of *calcitrans*; *occidentis* Walk. and *parasita* Fabr. are expressly stated to have plumose antennae, and must therefore belong to some other genus. As to *S. cybira* Walk., Walker himself questions its position in this genus. *S. calcitrans* L. is a brownish gray fly; its thorax has three rather broad, whitish stripes; on each border

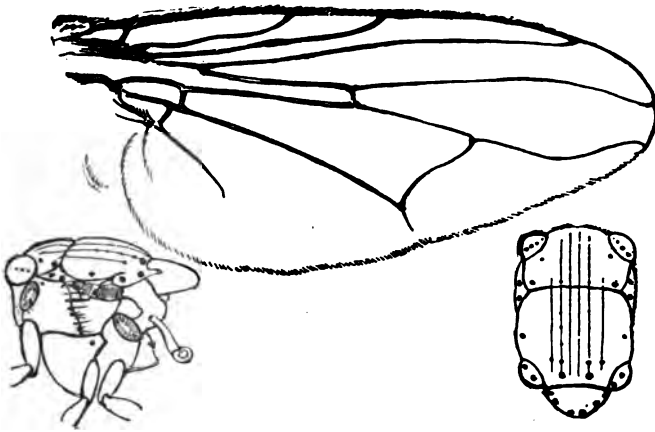


FIG. 1.

of the middle stripe and on the mesal borders of the lateral stripes is a blackish brown line; abdomen yellowish brown; on the second, third, and fourth segments are three brown spots which may be faint or even absent; wings hyaline or tinged with brown at base and along the costa. It has seemed to me that specimens taken on the borders of woods are more likely to have the brownish wings. Antennae brown; palpi yellowish brown; legs blackish brown, with yellowish or reddish knees.

Haematobia. — Two American species are known: *H. serrata* Desv. (Fig. 2 a), the "Horn Fly," an unpleasant importation from southern Europe, and *H. alcis*. Snow (Fig. 2 b), found by Professor Dyche in the cranberry swamps of northern Minnesota.

The two species are easily separated by the following points: hind tarsi of male serrate in *serrata*, not so in *alcis*; palpi black in *serrata*, yellow in *alcis*; pile of bucca black in *alcis*, yellow in *serrata*; at cephalo-dorsal angle of mesopleura *alcis*

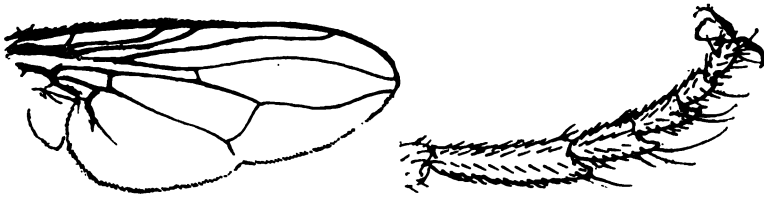
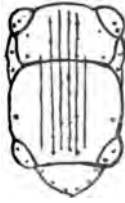


FIG. 2 a.



has a large macrochaeta curved dorsad; *serrata* has no macrochaeta at this point; at the cephalo-ventral angle of the mesopleura (protecting the pro-

stigma) *alcis* has two small bristles; *serrata* has a tuft of golden yellow hairs; *serrata* has much longer and denser pile on the dorsum of the thorax. Other color differences are pointed out by Professor Snow in connection with his description of *alcis* in *Can. Ent.*, April, 1891. I am much indebted to the Entomological Department of Kansas University for a pair of specimens of *H. alcis* which have enabled me to ascertain the important differences in the chaetotaxy of the mesopleurae above mentioned.



FIG. 2 b.

Hemichlora.—Only known species and type of genus *H. vittigera* Bigot, Mexico. Professor Williston, in his *Manual of North American Diptera*, p. 143, suggests parenthetically that this may be the *Idia viridis* of Wiedemann (*Auss. Zweif.*, Vol. II, pp. 354, 11). I quite agree with this suggestion for the following reasons. Meigen founded the genus *Idia* in 1826, and in his characterization (*Syst. Besch. Eur. Zweif.*, Vol. V, p. 9) the only distinguishing character is the pectinate arista. To this Wiedemann (*loc. cit.*, p. 347) adds: face prolonged forwards

below and entirely without hair. Wiedemann's description of *Idia viridis* is based on one poorly preserved and greasy specimen and reads: "With black antennae, everywhere bronzy green, with hyaline wings and blackish legs. Face and front rusty brown, the green color dark, tending toward emerald green."

Hemichlora has the pectinate arista, a slightly prominent oral margin, and is in part of a metallic blue color. A metallic blue color may vary to metallic green. Certainly *Hem-*

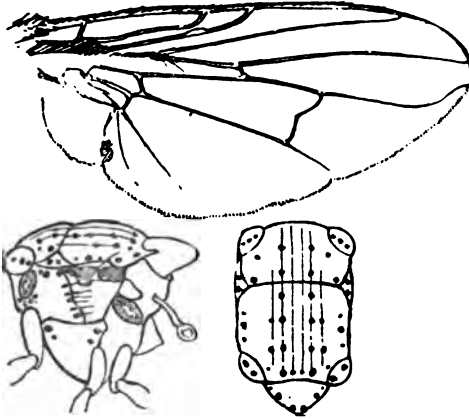


FIG. 3.

ichlora vittigera comes nearer to the description of *Idia viridis* than any other known North American Muscid. No other *Idia* has been described from North America, and only the original specimen of *I. viridis* is known.

Myospila.—There is but one known North American species, *M. mediotabunda* Fabr. (Fig. 3). It is common to Europe and America. Many of our specimens have the pubescence of the eyes very short; in the females sometimes it is very difficult to make

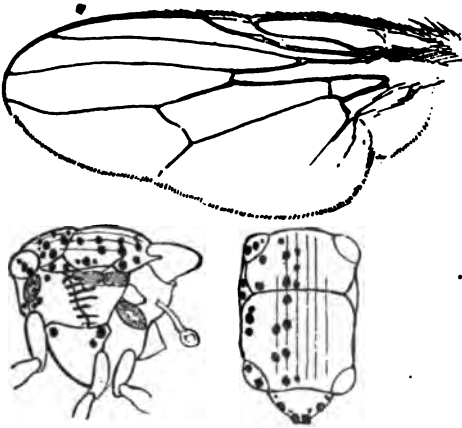


FIG. 4.

out with an amplification of twenty diameters. No other difference am I able to find between European and American specimens. It seems to me very probable that *Cyrtoneura quadrisetosa* Thomson (*Eugen. Resa*, p. 549) is one of those

specimens whose eyes are almost bare. I have seen such specimens from California.

Muscina.—Until within a few years our species of *Muscina* have been referred

to *Cyrtoneura*. Such authorities as van der Wulp and Williston are of the opinion that *Cyrtoneura* is a badly conceived genus

and that the name should be dropped, the species formerly referred to it being divided among the genera *Muscina*, *Clinopera*, *Hemichlora*, and *Morellia*. Similarly, *Cyrtoneurina* should be dropped as a generic name, its species being distributed among

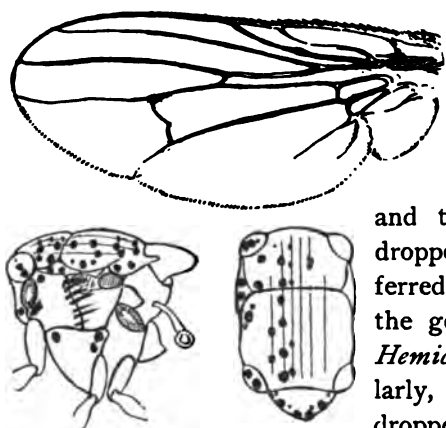


FIG. 5.

some of the genera just mentioned. Following van der Wulp's views in cases where the species are unknown to me, the following are the known North American species of *Muscina*: *stabulans* Fall., *assimilis* Fall., *mexicana* Macq., *pallidicornis*

Bigot, *parilis* Gigl.-Tos, *vecta* Gigl.-Tos, *linea* v. d. W., *tripuncata* v. d. W., *aurantiaca* nov. sp., and *texana* nov. sp.

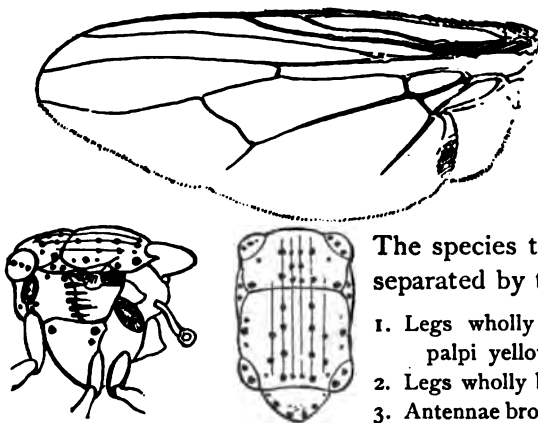


FIG. 6.

The species that I know may be separated by this table:

1. Legs wholly or partly yellow;
palpi yellow 3
2. Legs wholly black 4
3. Antennae brown; three pairs acrostichal
bristles cephalad the transverse suture; prostigma brown;

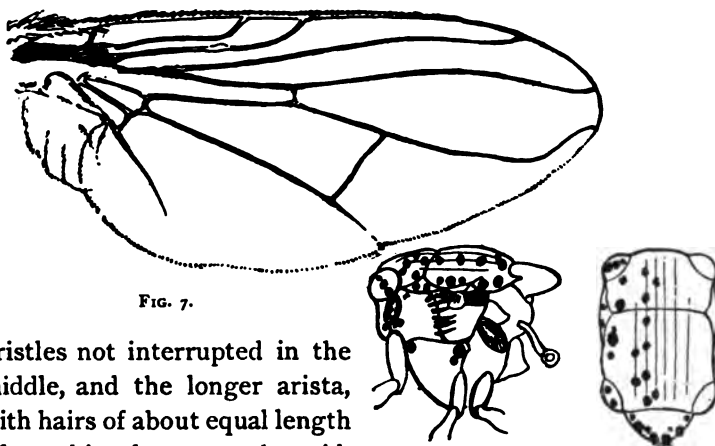
humeri concolorous with thorax *stabulans* Fall. (Fig. 4)

Antennae pale yellow; one pair acrostichals cephalad the suture; prostigma whitish yellow; humeri not concolorous with thorax *texana* nov.sp. (Fig. 7)

4. Palpi and antennae *assimilis* Fall. (Fig. 5)
 Palpi and antennae orange yellow . . . *aurantiaca* nov. sp. (Fig. 6)

M. aurentiaca nov. sp. — Male and female; several specimens collected by Mr. G. R. Pilate at Tifton, Ga. General appearance like that of *stabulans*, *assimilis*, and *pabulorum*, i.e., with gray-striped thorax and abdomen with variable spots. Chaetotaxy like *stabulans*, etc.

M. texana nov. sp. — Two males, Texas. Agrees very closely with Giglio-Tos's descriptions of *M. parilis* and *M. vecta*, from which its rather broad front, with the series of transfrontal



bristles not interrupted in the middle, and the longer arista, with hairs of about equal length (if anything longer at the middle of the arista than at the base), and much more scattered than in *vecta* and *parilis*, clearly separate it. The same characters separate it from *M. linea* v. d. W. I separate it from *tripunctata* v. d. W. because of the striking appearance of the prostigma, which I think Mr. van der Wulp would hardly have failed to note had it been present in his specimens.

Clinopera (Fig. 8, *C. inuber* G.-Tos). — Mr. van der Wulp describes in *Biologia Centrali Americana* seven species, and refers to this genus *Cyrtoneurina uber* G.-Tos, *inuber* G.-Tos, *gluto* G.-Tos, and *pellex* G.-Tos. All these species are Mexican. He also refers *Cyrtoneura anthomydea* Bigot to *Clinopera*. It seems to me that there is nothing in Bigot's description that does not apply to *Muscina assimilis* Fall., specimens of which

from the Rocky Mountains (the source of Bigot's specimen) are in my collection.

MUSCINAE MUSCAEFORMES.

1. Middle tibia with a prominent macrochaeta on its flexor surface . . . 2
(The male of some European Mesembrinae has no such macrochaeta, but its middle tibiae are elongate and on their flexor surface thickly hairy.)

Middle tibiae without such a prominent macrochaeta . . . 3

2. Sternopleural macrochaetae 1 : 2 ; elbow of fourth vein forming a rounded angle ; no orbital macrochaetae, but the geno-vertical plate uniformly beset with minute bristly hairs ; transfrontal macrochaetae small and weak, often difficult to see . . . *Pseudopyrellia* Girschner.

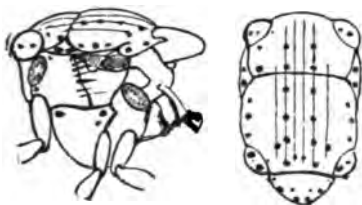


FIG. 8.

Sternopleural macrochaetae 1 : 3 ; first longitudinal vein ending in the costa at about the middle of the wing ; small cross-vein about opposite the end of the first longitudinal ; elbow of fourth longitudinal not forming an angle but a gentle curve convex toward the hind border of the wing forming

an apical cross-vein which is longer than the terminal portion of the fourth vein before its curvature . . . *Pyrellia* Desvoidy

Sternopleural macrochaetae varying, 1 : 3, 1 : 2, 0 : 1 ; if (rarely) 1 : 1 then the anterior notably the smaller ; first longitudinal vein ending in the costa far beyond the middle of the wing ; small cross-vein a long distance basad to the end of the first longitudinal ; fourth longitudinal sweeping in a broad curve, convex toward hind border of wing, towards the third, thus forming an apical cross-vein, which, however, in our species, is not longer than the terminal portion of the fourth vein before its curvature . . . *Mesembrina* Macquart

3. Bend of fourth longitudinal forming a rounded angle. Outline of arista as a whole fan-shaped. Sternopleural macrochaetae 1 : 2.

Musca Linnaeus

Bend of fourth longitudinal not forming an angle at all, but a gentle curve . . . 4

4. Antennae separated at their base by a distinct ridge ; sternopleural macrochaetae 0 : 2 ; eyes very distinctly hairy *Graphomyia* Desvoidy
Antennae not thus separated ; sternopleural macrochaetae 1 : 2 ; eyes not distinctly hairy . . . 5
5. Arista naked . . . *Synthesiomyia* Brauer and Bergenstamm
Arista plumose . . . *Morellia* Desvoidy

Pseudopyrellia (Fig. 9).—Our only known species is *P. cornicina* Fabr. I consider *Lucilia carolinensis* Desv., *L. compar* Desv., and *L. Heraea* Walk., as synonyms.

Pyrellia (Fig. 10).—The only North American species that I have seen is *P. cyanicolor* Zett. The specimens agree perfectly with the description and with European specimens from Prof. G. Strobl and others. *P. setosa* Lw. is a synonym; I have compared the types in the Agassiz Museum with my

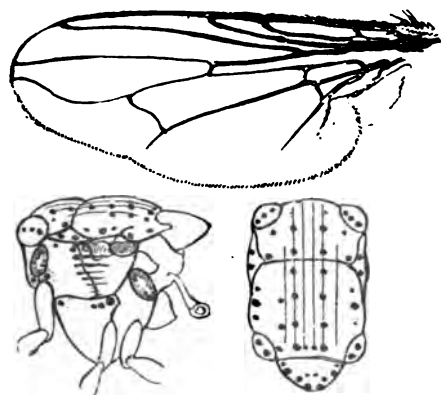


FIG. 9.

American and European specimens. Occasionally one finds a specimen which is of a rather bright metallic green instead of the usual dark steely blue, but I can find no structural or other color differences. *Musca occidentis* Walk., *Dipt. Saund*, p. 347, is probably this same species.

P. cadaverina L. may be distinguished from *cyanicolor* by the entire absence of any trace of hoary coating on the thorax even at the cephalic border, while *cyanicolor* has three broad hoary stripes, a median and two humeral, which are specially distinct at the cephalic border.

Mesembrina.—There is but one known North American species, which we must call *M. latreillii* Desv. (Fig. 11) because no other species is known. Desvoidy says: "Tout à fait semblable au *M. meridiana*; un peu plus petite; antennes brunes; la face est d'un argenté brillant sur les côtés." Now

FIG. 10.

the species is by no means just like *M. meridiana* (Fig. 12); it does not average smaller than that species; the antennae vary in color from yellow to brown; the sides of the face are, however, silvery. The species agrees perfectly with the description of

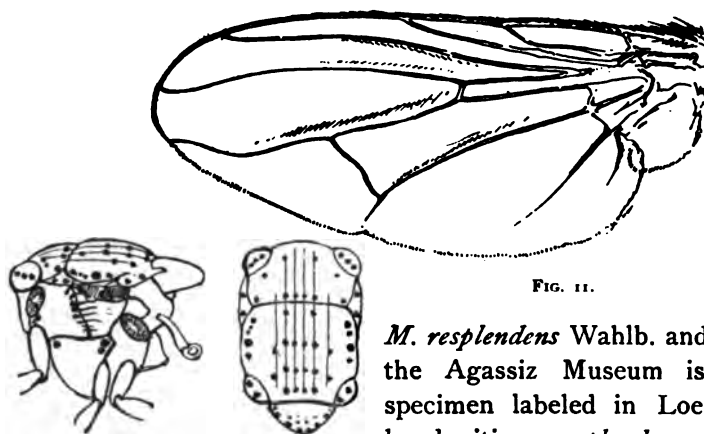


FIG. 11.

M. resplendens Wahlb. and in the Agassiz Museum is a specimen labeled in Loew's handwriting *resplendens*. I

think there can be no doubt of the synonymy. Mt. Washington, N. H.; North Mt., Pa.; Seattle, Wash.; Dakota. For comparison I introduce here Fig. 13, *M. mystacea* L. If the truth could ever be known, it is highly probable that *M.*



FIG. 12.

pallida Say would be found to be an *Oestrophasia* very near or identical with *O. punctata* Coq.

M. anomala Jaennicke is evidently not a *Mesembrina* at all. Professor Brauer suggests that it belongs near *Spilogaster*.

Musca. — *M. domestica* L. (Fig. 14) is very common. Walker says that *M. corvina* Fabr. occurs in Nova Scotia. The front of *corvina* is very narrow in the male, that of the male *domes-*

tica is about one-fourth as wide as the head; the frontal vitta of the female *corvina* is of uniform width throughout, while that of the female *domestica* is narrow near the antennae and broadens out very considerably towards the vertex. Specimens from



FIG. 13.

Jamaica, agreeing with Macquart's description of *M. basilaris*, are certainly *M. domestica*.

Graphomyia (Fig. 15). — Our single species does not differ structurally and has but insignificant color differences from *G.*

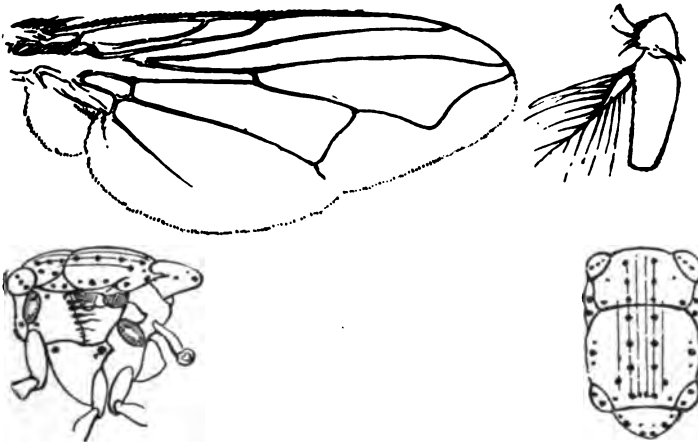


FIG. 14.

maculata Scop., with which I identify it. *G. americana* Desv. is the same species.

Synthesiomyia. — *S. brasiliiana* BB. occurs in Florida and in Georgia (Fig. 16). It is the only species of the genus.

Morellia. — *M. micans* Macq. (Fig. 17) is very common all over the United States. Bluish black; thorax with three broad

hoary stripes; last abdominal segment of female brown with hoary coating. Macquart described the female only; the male resembles the female in color (except that the last abdominal

segment has much less of a brown color) and has the following structural features which are characteristic: a

beard of long hairs on the mesal border of the hind tarsi and a fringe of short, fine hairs on the anterior surface of the middle tibiae not far from the extensor border.

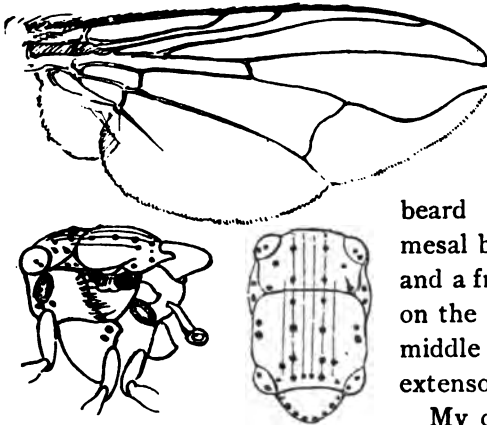


FIG. 15.

My collection contains also two species of *Morellia* from

Mexico and Jamaica. One is rather variable, the variations corresponding to the descriptions of *Musca violacea* (Fig. 18) Fabr., *Pyrellia maculipennata* Macq., *P. specialis* Walk., *P. sus-*

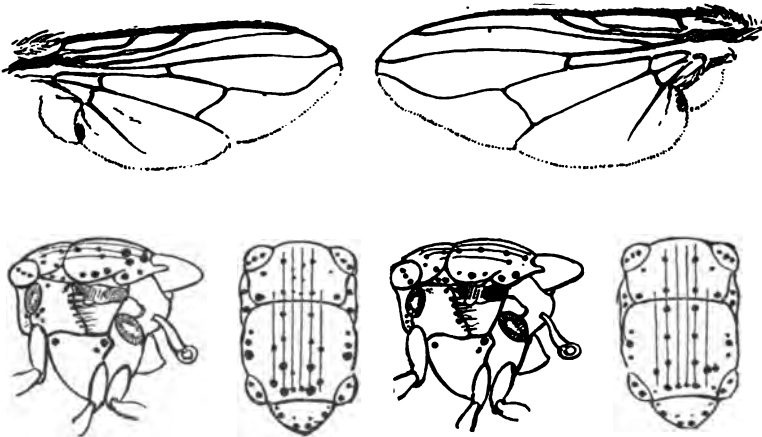


FIG. 16.

FIG. 17.

picax Walk., *P. basalis* Walk., *P. centralis* Lw. (types compared and found to agree), and *P. iris* Bigot. Fabricius's name has priority. The male has on the anterior surface of

the middle tibia, near the extensor border, a series of bristles extending from base to apex; the basal half of the series are very tiny but stout as compared with their length, real little spines; at about the middle of the series the form changes gradually to that of a delicate and moderately long bristle. This arrangement is very like that of the European *M. hor-torum* Fall. My specimens of this species are from Jamaica, C. W. Johnson, and from Mexico, O. W. Barrett.

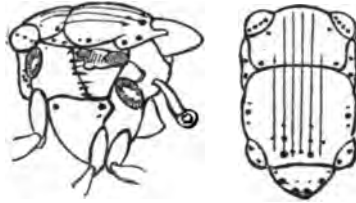


FIG. 18.

The other species agrees with the descriptions of *Pyrellia scapulata* Bigot (Fig. 19) and *P. flora* Bigot. The middle tibia

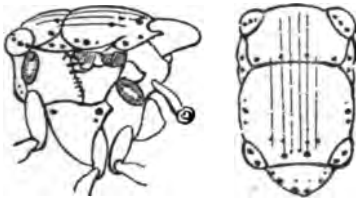


FIG. 19.

of the male has some noteworthy structures. On the anterior surface at the base is a tubercle about 0.3 mm. (one-seventh the length of the tibia) long and half as broad, its long diameter parallel to that of the tibia. On this tubercle is a row of six or eight very short thick spines, and in some specimens a second row, flexad the first, of about four similar but much smaller spines, can be made out. From the apical end of the tubercle the principal row is prolonged as a series of about equal and equidistant, much more delicate spines, as far as the apex of the tibia. Just at the junction of the posterior and flexor surfaces there are three large bristles; one is at the junction of the basal and middle thirds, the second at the middle of the tibia, the third at the junction of the middle and apical thirds. My specimens of this species are from Jamaica, collected by C. W. Johnson. The name *scapulata* has priority.

CATALOGUE OF MUSCINAE MENTIONED IN THIS PAPER BUT NOT
REFERRED TO IN OSTEN-SACKEN'S CATALOGUE.

- Haematobia serrata* Desv., Myod., 389, 3.
Haematobia alcis Snow, *Can. Ent.*, April, 1891. Vol. xxiii, pp. 87-91.
Hemichlora vittigera Bigot, van der Wulp, *Biol. Cent.-Amer.* Vol. ii,
 p. 304.
Cyrtoneura Bigot, *Bull. Soc. Zool. Fr.*, p. 613, male. 1887.
Cyrtoneurina Gigl.-Tos, *Mem. R. Accad. Sci. Torino.* Ser. 2, vol.
 xlv (sep.), p. 13, female.
Muscina assimilis Fall.
Musca assimilis Fall., *Dipt. Suec.*, *Musc.*, 56, 41. 1820.
Musca caesia Meigen, *Syst. Besch.* Vol. v, p. 76, 43. 1826.
Muscina concolor Desv. (?), *Myodaires*, p. 408, 5. 1830.
Muscina fungivora Desv., *loc. cit.*, p. 408, 6.
Cyrtoneura aperta Macq., *Dipt. du Nord de Fr.*, 11, 4. 1834.
Musca borealis Zett., *Ins. Lapp.*, p. 660, 28. 1838-40.
Cyrtoneura caesia Meig., *Syst. Besch.* Vol. vii, p. 309, 5. 1838.
Cyrtoneura aperta Macq., Meigen, *loc. cit.* Vol. vii, p. 309, 14.
 1838.
Cyrtoneura caesia Meig., Zetterstedt, *Dipt. Scand.* Vol. iv, p. 1351,
 5. 1845.
Cyrtoneura assimilis Fall., Zett., *Dipt. Scand.* Vol. iv, p. 1351,
 6. 1845.
Cyrtoneura caesia Meig., Schiner, *Fauna Austr.* Vol. i, p. 597.
 1862.
Cyrtoneura assimilis Fall., Schiner, *loc. cit.*, p. 598. 1862.
Cyrtoneura assimilis Fall., Rondani, *Dipt. Ital. Prod.* Vol. v, pp. 214
 and 216. 1862.
Cyrtoneura assimilis Fall., Strobl, *Dipt. Steiermark.* Vol. ii, p. 76.
 1894.
Muscina pallidicornis Bigot, van der Wulp, *loc. cit.*, p. 311.
Cyrtoneura pallidicornis Bigot, *Bull. Soc. Zool. Fr.* Vol. xii, p. 614.
 1887.
Muscina parilis Gigl.-Tos, van der Wulp, *loc. cit.*, p. 311.
Cyrtoneurina parilis Gigl.-Tos, *loc. cit.*, p. 14, No. 154.
Muscina vecta Gigl.-Tos, van der Wulp, *loc. cit.*, p. 311.
Cyrtoneurina vecta Gigl.-Tos, *loc. cit.*, p. 14, No. 155.
Muscina lineæ v. d. W., *loc. cit.*, p. 304.
Muscina trilineata v. d. W., *loc. cit.*, p. 305.
Muscina texana nov. sp.
Muscina aurantiaca nov. sp.
Clinopera. Mr. van der Wulp's species are described in *Biol. Cent.-Amer.*,
Dipt. Vol. ii, pp. 305-310. He includes *C. uber* Gigl.-Tos and

inuber Gigl.-Tos in his table, p. 306, and makes some remarks on them, pp. 307 and 308. On p. 311 he refers *Cyrtoneurina gluto* Gigl.-Tos and *pellex* Gigl.-Tos to *Clinopera*. All these species of Gigl.-Tos were described as *Cyrtoneurinae* in Mem. R. Accad. Sci. Torino, ser. 2, vol. xlv (sep.), pp. 14-17. *Cyrtoneura anthomydea* Bigot, Bull. Soc. Zoöl. Fr., vol. xii, p. 614 (1887), is referred by Gigl.-Tos, *loc. cit.*, p. 15, to *Cyrtoneurina*, and by van der Wulp, *loc. cit.*, p. 311, to *Clinopera*.

Pyrellia cyanicolor Zett., Dipt. Scand. Vol. iv, p. 1323, 4.

Pyrellia serena Meig., Rondani, Dipt. Ital. Prod. Vol. v, p. 203.

Pyrellia cyanicolor Zett., Strobl, Dipt. Steiermark. Vol. ii, p. 73.

Pyrellia setosa Loew, Centuriae. viii, 63.

Synthesiomyia brasiliiana BB. Brauer and Bergenstamm, Vorarb. z. Monog. Musc. Schiz. Vol. iii, pp. 96 and 110.

Morellia violacea Fabr.

Musca violacea Fabr., Syst. Antl., p. 288, No. 25; Wied., Auss. Zweif. Vol. ii, p. 409, 43.

Cyrtoneura violacea Fabr., Brauer and Bergenstamm, *loc. cit.*

Pyrellia maculipennata Macq., Dipt. Exot. Supp. 4, 252, 12.

Cyrtoneura maculipennata Macq., Townsend, Ann. New York Acad., p. 33. 1892.

Pyrellia iris Bigot, Ann. Soc. Ent. Fr., p. 36, female. 1878.

Pyrellia centralis Loew, Centuriae. viii, 62.

Morellia scapulata Bigot.

Pyrellia scapulata Bigot, *loc. cit.*, p. 35, female. Mexico.

Pyrellia flora Bigot, *loc. cit.* Male. Haiti.

EXPERIMENTAL STUDIES UPON HYDROMEDUSAE.

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IN connection with previous work on regeneration the problem of animal grafting was repeatedly suggested by many phenomena associated with that work, and during the summer of 1898 a series of experiments was undertaken at the Marine Biological Laboratory and carried on through the months of July and August.

It is a pleasure in this connection to acknowledge many courtesies extended by the director, Prof. C. O. Whitman, and valuable suggestions from Dr. Jacques Loeb, during the progress of the work. The following paper aims simply to present a résumé of methods and results, deferring speculative considerations which might be suggested by any of the phenomena involved.

I. *Material.*

This was restricted to two sorts of organisms, Hydroids and Medusae. Of Hydroids an almost unlimited amount and of many genera and species were available and easily obtained from the waters about the docks of the U. S. Fish Commission, from the rocks and fucus in the harbor and adjacent waters. It was obtained fresh every morning and experiments were made only upon it within a few hours. The genera used in the experiments were Eudendrium, Pennaria, Parypha, and a few of the Campanularidae.

Of Medusae only one species was available in sufficient abundance and size for experimentation, namely, *Gonionemus vertens*, a Medusa occurring in considerable numbers in the "eel pond," a small body of water adjacent to the laboratory and communicating with the waters of the harbor by a very

narrow inlet. As this Medusa endures artificial conditions with considerable ease for days and even weeks, it lends itself readily to such experiments. Its size also, varying from 2 or 3 mm. to as many cm., is also a factor of convenience, though a larger species would prove desirable in some operations. Its activity during at least three months also facilitates extended experiments.

II. *Methods.*

In grafting the Hydroids, the stems were cut into fragments varying from 5 to 10 mm. in length, and usually taken from the younger and fresher portions of the stem. Successful unions were made from older portions but in much smaller proportions, and requiring much longer time. The hydranths were excised, since the motion of the body and tentacles would invariably disturb the contact of the specimens. Having prepared suitable sections, they were brought into contact in watch-glasses, or small petri dishes, of perfectly fresh sea water, and retained in position by small bits of lead shaved freshly from thin sheets. Bits of platinum wire would have been better, though it was not available at the time, and little appreciable difficulty was had with the lead. The water was changed on the specimens daily.

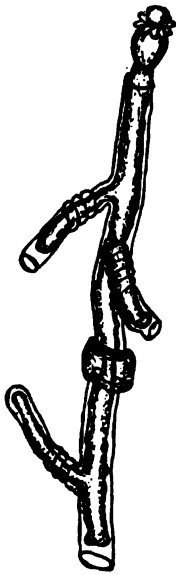


FIG. 1.

With the Medusae the task was a much more difficult one, for while the power of spontaneous movement had been largely eliminated by emarginating the bell and thus removing the marginal nerve ring, and thus the centers of spontaneity, the contractility had not been destroyed, and for a time great difficulty was found in retaining the parts in even contact. And in this connection I desire to correct a partial error in my previous paper on "Regeneration,"¹ where I had failed to recognize the paralyzing effects of such emargination, a fact due to failure

¹ *Zoölogical Bulletin*, I, p. 29.

to remove sufficient of the margin. If the least portion was left, or even a portion quite near the margin, automaticity was retained, but by removing carefully and completely some 2 or 3 mm. of the bell-margin I was able to confirm quite fully the experiment of Romanes¹ on Scyphomedusae.

After various expedients had failed, resort was had to bits of rather fine shoemaker's bristles, cut into proper lengths. These were thrust through the gelatinous portions of the Medusae in such directions as would serve to fairly secure the desired contact of the surfaces to be united. An inspection of the several figures will give the best idea of how this was secured. Cf. Figs. 8 and 9. But at best the method was only partially successful. It may be mentioned in this connection that one of the difficulties attending these experiments was the danger of deleterious contamination involved in the whole of

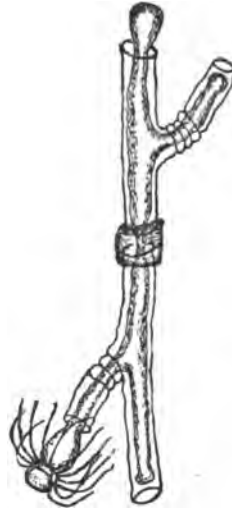


FIG. 2.

the operations from Bacteria and parasitic Infusoria invading the water and impeding or destroying the experiments. This was much more noticeable in experiments on Medusae than on Hydroids, a fact due in part, certainly, to the promptness with which the latter reacted in regeneration of tissues as compared with the former. Notwithstanding, it is rather remarkable that so small a proportion in either case suffered, since no *special* pains were taken in the way of critically guarding against contamination beyond the more ordinary provisions of clean glassware and instruments. To maintain a fairly equable temperature during unusually hot days, the covered vessels containing the specimens were set under the running water taps of the laboratory.

¹ Jellyfishes, etc., p. 27 *et seq.*

III. Grafting.

Hydroids. — The work upon Hydroids was restricted chiefly to species of *Eudendrium*, *Pennaria*, and *Parypha*, though a few experiments were tried upon *Campanularians*. Upon the latter the results were almost entirely *negative*, though for this no apparent cause was ascertained. The experiments were not of sufficient numbers, nor of sufficiently varied conditions, to warrant any conclusions as to the incapacity of these to regenerate or coalesce. Time was not adequate to extend the

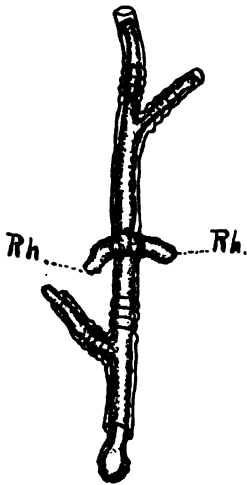


FIG. 3.

attention to this group which might have resulted more favorably. Again, the relatively small size of the members of this group available was a further embarrassment to successful experimentation. However, neither of these is offered as sufficient account of the negative character of the experiments. Indeed, the work of Davenport ('94) on *Obelia* is strongly affirmative, at least so far as regeneration is concerned.

Experiments upon species of the genera named were specially successful. While of course in all such work a large number of failures must result, yet when the difficulties of manipulation and the

artificial conditions necessary are considered, this is not strange.

While no mathematical estimates were made as to the ratio of successful experiments, I think it may be safely said that at least 20 per cent of all were successful. It need hardly be pointed out that results varied materially in both the time necessary, and the degree of perfection, in the coalescences. This will be noted in detail in connection with the several experiments described.

A comparison of the several figures will perhaps indicate in general better than words the methods and results. Cf. Figs. 1-6.

The union between sections of the same species was usually quite perfect within from eighteen to thirty-six hours. A

delicate sheath of perisarc secreted over the ends was the first indication of special activity and regeneration. This usually occurred at any wounded point. It became specially marked at the points of contact of the grafted specimens. The first effect of the sectioning of the specimens preparatory to their being placed in contact was a pronounced contraction of the coenosarc within the tubular perisarc, and the closure of the cut ends of the enteric cavity. This was usually, however, soon followed by an outgrowth till the coenosarc of the two specimens came into contact, when the secretion of the extra perisarc proceeded as a joint operation, though sometimes by a single one, if its activity and response were the more prompt. *Cf.* Figs. 1 and 2.

Following this the contact of the two became more intimate, the healed ends united with each other, fusion being followed by the absorption of the terminal portions and the consequent confluence of the enteric channels and their contents.

In the experiments no apparent difference was noticeable as to anything like polarity, the parts uniting orally, aborally, or otherwise, with equal freedom and promptness. With species of *Eudendrium* and *Pennaria* this was demonstrated with absolute certainty, the directions of the branches making any mistaking impossible. *Cf.* Figs. 2-4.

In these species the sexes are distinct, and experiments in grafting specimens of the opposite sexes were quite as prompt and perfect as otherwise. There would seem, therefore, to be not only no definite differences of polarity as seems to be the case in *Hydra*, but no sexual difference in so far as regenerative or coalescence capacity is concerned. It remains to note results as to grafting different species. With none of these are the distinctions sufficiently clear to warrant positive con-

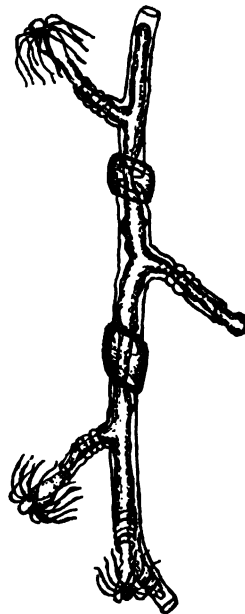


FIG. 4.

clusions. Of *Pennaria*, only one species was available, and the same was true of *Parypha*. Of the species of *Eudendrium* there have been several indicated, but their distinctness is to my mind open to serious doubt.

If the distinctness of Agassiz's species of *E. dispar* and *ramosum* is to be maintained, then the grafting of these has been as clearly established as that of the different sexes. It would not be strange should closely allied, though definitely distinct, species be found to coalesce in these organisms, for such has been long known among plants, and shown for animals by the recent experiments of Born ('96), Crampton ('97), Harrison ('98), and others. But so far as I am aware it has not hitherto been demonstrated for the Hydroids; indeed, most of such efforts have been negative in results.

As to the coalescence between specimens of different genera, the experiments seem to be conclusive and wholly negative. Out of a considerable series, while there were indications of temporary union, in no case did it become con-

clusively permanent. The most favorable indications were upon *Eudendrium* and *Pennaria*, Hydroids of very similar size, structure, and habit; but after repeated experiments under different conditions the results were as already indicated. In Fig. 6 it will be noted that the usual secretion of perisarc at the points of contact has been deposited, and apparently by the coöperation of both sections; still at no time were there evidences of organic union of the coenosarc, and later this was distinctly withdrawn, and the sections continued an independent existence, each producing new hydranths, though

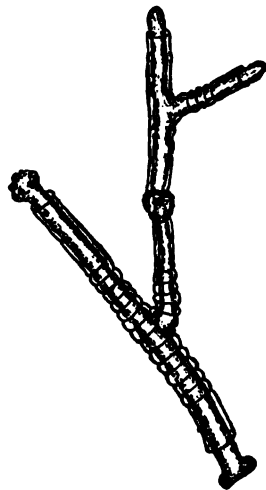


FIG. 6.

in the case of *Eudendrium* they continued rudimentary in the specimen figured.



FIG. 5.

IV. *Medusae.*

Experiments upon Medusae were restricted to *Gonionemus vertens*, being the only species available in sufficient numbers, and capable of adaptation to the artificial condition necessitated by the nature of the work. This Medusa is found in great numbers, though only in a limited locality adjacent to the Marine Laboratory, and lives readily for several days, or even weeks, in table aquaria, if reasonable precautions be taken to keep the water fresh and supply suitable food.

While the experiments were quite extensive and various, aiming at first to ascertain the trend of resultants, no attempt will be made in this connection to describe them in any con-



FIG. 7.



FIG. 8.

siderable detail, but rather to call attention to a few of the more conspicuous of them, and to indicate something of their probable significance.

Fig. 7 will afford a good idea of the general features of the Medusa with the entire margin of the bell and its organs excised, preparatory to any contact experiments.

In Fig. 8 are shown two Medusae from which portions have been removed, the larger part of each being brought into contact and retained by the bristle, *br.*, passing through the body. In some instances two or three bristles were passed through in different planes, thus giving greater stability of contact.

Parts of various sizes and from different regions were similarly grafted, and with usually similar results. The time required for union differed greatly in experiments conducted under exactly the same conditions and care. In some cases complete union had taken place within twenty-four hours, while in others it only occurred after several days. It should be noted, however, that when once coalescence had begun it usually went forward with comparatively great rapidity.

In Fig. 9 is shown the coalescence of two Medusae by their oval margins. This was usually the most easily performed of any of the experiments upon the Medusae, and union was rather more prompt if any difference was noticeable. As will be seen from the figure, fusion was not entire in this case, a

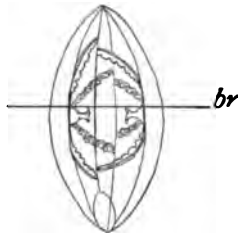


FIG. 9.

small mouth-like opening remaining on one side through which aëration of the interior was made possible, and by means of which by contraction the united individuals were able to move about in the water. It should be noted in this connection that upon stimulation coördinated movements were produced, and in a few instances even apparently spontaneous movements were clearly recognized. I have said that this action was *apparently* spontaneous. It is not impossible, of course, that some extraneous stimulus might have been involved, but, if so, it was wholly beyond any ordinary physical detection, and was distinctly recognized by several persons to whom it was pointed out.

In these experiments, in a few cases, the specimens united completely throughout the entire margin, but with the result that the specimen died within a comparatively short time, presumably from inability to secure aëration of the portions where metabolism was most active and aëration most imperative.

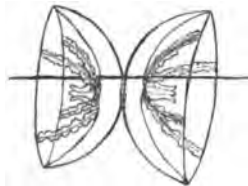


FIG. 10.

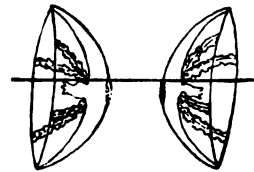


FIG. 11.

Figs. 10 and 11 show a method of aboral grafting made upon a considerable number of specimens, but uniformly without permanent success. As will be noted in Fig. 11, the points of contact were slightly scarified, or in some cases portions excised with sharp scissors, in order to favor coalescence of

the surfaces, but after a few hours the specimens would almost invariably have drawn apart by some sort of creeping movements, probably aided in part by the prehensile character of the manubrium, and usually one or both finally extracting the bristle entirely. I am not able to suggest any satisfactory explanation of the negative character of this experiment. Whether regenerative tissue is wanting on this area, or whether some intrinsic repugnance to such fusion be the cause or occasion, or whether some cause wholly undetected was present, seems a matter of doubt. There would seem to be no *a priori* reason why this particular experiment should not find as ready a response as those already described. The inverted position could hardly be assigned, for specimens in similarly inverted aspects readily united by the margins, as indicated in Fig. 12.

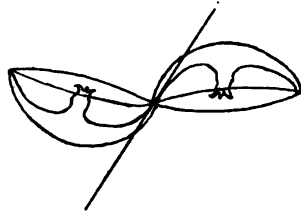


FIG 12.

In Figs. 13-16 is shown a phenomenon which appeared in connection with the series of experiments quite incidentally, and of which I shall undertake no particular explanation, and yet which is one of the most novel and interesting of the entire series.



FIG. 13.



FIG. 14.



FIG. 15.



FIG. 16.

In certain aboral grafts, similar to those already described in Fig. 10, a single specimen was found with the bell somewhat evaginated, as in Fig. 13. During the following days, July 27 and 28, it passed successively through the phases represented in Figs. 14, 15, and 16, becoming permanently united in the completely evaginated form of Fig. 16, in which condition it continued to live and even take particles of food, though it showed no evidences of growth, beyond a distension of the gastric pouch due to the engulfed food. Finally, on August 3 the specimen died during the very sultry night, and was found the

following morning in a partially disintegrated condition, a result due in part to the unusual temperature, and perhaps in part to overfeeding.

A series of experiments were undertaken by which to secure, if possible, by artificial means other inversions of a similar sort, and though various expedients were adopted by which to facilitate such, they gave no permanent results.

The phenomenon of eversion in the Medusae of *Obelia* and other Hydroids in their young or newly discharged condition is quite well known, but it continues for a brief time only, and with no disposition toward permanence so far as I have known.

Of course the classical experiments of Trembley (1744), Nussbaum ('87, etc.), and others upon *Hydra* are too well known for special comment, and at first sight might be thought to be analogous to that now under consideration; but a moment's reflection will suffice to show that it is only so in a very general way. For example, there is no inversion of the relative position of ectoderm and endoderm, since the lining of the bell, outer surface of the manubrium, etc., is ectodermal. The enclosed cavity of the everted specimen served no new function in its changed condition, nor did the outer layer in its new relations. That any change in the histological characters would be induced may therefore be considered very unlikely since the change of relative position, while considerable, is yet not such as would necessitate any change of function. Nevertheless the fact is an interesting one, and apparently quite unique. At one time, just about the completion of union of the inverted margins, a decided papilla-like bud appeared at the aboral area which presented some resemblance to a second manubrium, but this soon after was absorbed entirely, and was probably only the elevation due to the approximation of the margins preparatory to final union.

V. *Regeneration and Heteromorphosis.*

In connection with the foregoing experiments occasion was taken to repeat some of my earlier experiments on regeneration and to extend them somewhat. At an earlier point in

this paper I have indicated an error in the former paper, as to the paralysis of the Medusa following the complete removal of the marginal portion of the bell. I desire, moreover, to express more definitely than appears in the earlier paper, though it was clearly implied at several points, the fact that in all those experiments there does not appear to be any actual increase of mass, or growth of the body as a whole, but that in all the regenerative activity it was evidently at the expense of other portions of the body proper. This would naturally follow in most cases, since in producing a new manubrium, or new tentacles, or in the grafting experiments, the animal was practically incapacitated for obtaining food, and under the artificial conditions of the experiment could hardly have been suf-

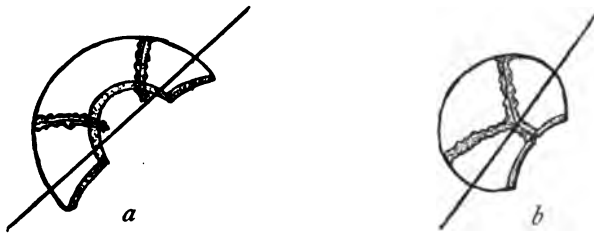


FIG. 17.

ficiently fed to make evident any growth. In many cases a very evident decrease in size was apparent in the progress of the experiment. Indeed, in many cases manubrium, velum, tentacles, etc., continued to live for weeks accompanied by a gradual decrease in the body mass, till it finally became wholly consumed, after which the organs gradually disintegrated.

In all essentials the later experiments confirm those earlier made. Further attempts were made to put specimens under such conditions as would render difficult any mere contraction or approximation of the surfaces. Figs. 17 and 18 will show two out of a considerable number and variety of the experiments. In these the portions of the body were set in their relative positions by bristles in such a way that only continued contractions of considerable vigor would be able to change them. But within forty-eight hours the results indicated in the several figures had taken place, the stereotype form of body assumed,

and without indication of specific regenerative growth and absolutely no hint at heteromorphism in the slightest way.

Figs. 19 and 20 show experiments designed to further test the regeneration of the manubrium. The operation of removing the organ was made on August 4. As will be seen in the case of Fig. 19, about three-quarters of the animal were excised, leaving one chymiferous canal fairly complete, and a mere remnant of a second, the whole of the manubrium hav-

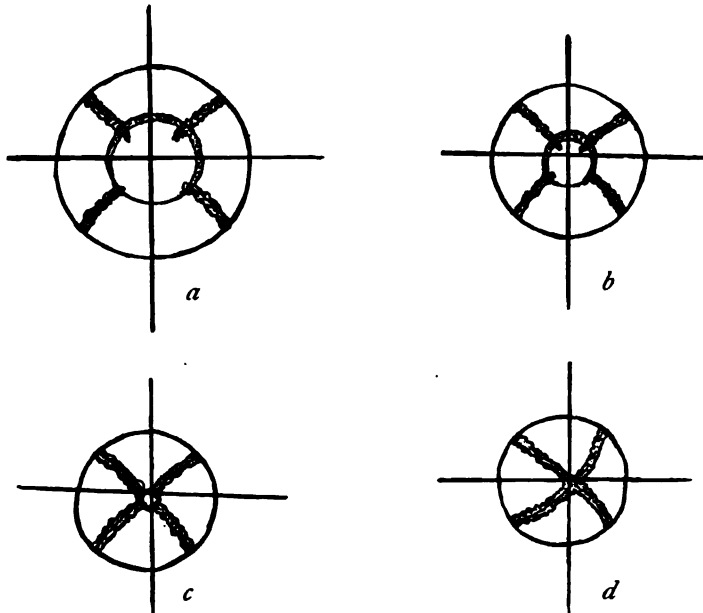


FIG. 18.

ing been removed, as indicated by the line of the cut, *a-b*. In Fig. 20 a wedge-shaped piece, about one-fourth the body mass, including the manubrium, one entire canal and the central portions of the other three, as indicated, was excised.

On August 5 the cut margins in each case had approximated each other and were evidently uniting. On August 7 the union was complete and the Medusae were swimming quite freely and naturally. On August 9 the first indications of a new manubrium were apparent, and in approximately the normal position. Its color and texture clearly indicated its forma-

tion as a new outgrowth, there being only the slightest traces of pigment present. The growth was quite gradual, and not until about August 14 had it become fully formed and functional.

In each case there had also been regenerated additional radial canals, as indicated in the figures. These appeared in connection with the lines of union, and were not at first

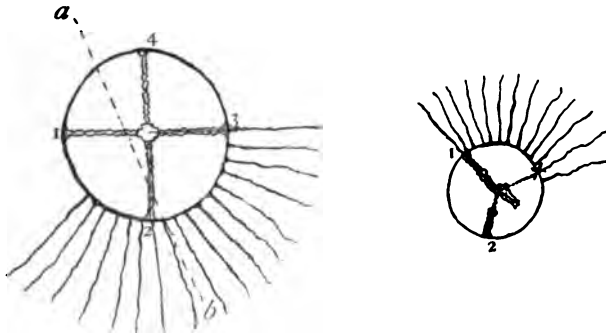


FIG. 19.

suspected as canals. Later the deposition of pigment along their course pointed strongly to the conclusion that they were canals, though whether yet functional I am not able to say.

In none of my experiments has there been any clear confirmation of the results and conclusions of Bickford ('94) that in Hydroid regeneration the polyp, tentacles, etc., are produced

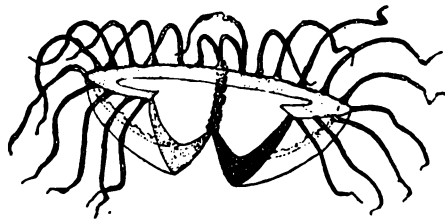


FIG. 20.

simply by a "transformation of the tissues of the stem"—that is, that the tentacles are formed by a sort of longitudinal cleavage of the coenosarc, and a remolding of it directly, without any tissue changes. In cases where there is only a recasting of a portion of the body into the *form* of the original, as in the case of a Medusa divided into sections, where each

part assumes the typical shape of the whole, being only different in *size*, this is of course clear. But from an inspection of Fig. 1 it will be evident that the tentacles are regenerated by a process of budding and growth exactly as in *Hydra*. The same was even more evident in the regeneration of tentacles, manubria, etc., in the *Medusae*. In every case they originate as minute buds, and become functional only after a considerable period of growth.

Whether additional tissue is formed from "*a few undifferentiated cells*," a sort of reserve embryonic tissue, may indeed be doubtful. Still, that there is *growth* in the ordinary histogenic sense must be evident in these cases as truly as in that of the regeneration of the tail or limb of the newt. And, indeed, it hardly seems to be more than a verbal quibble whether it be by one or *both* processes. For in either case it simply implies the presence in these organisms of cells or tissues possessing the capacity, shared in common with embryonic or undifferentiated tissues, of reparation.

VI. *Historical.*

Experimental work on the Hydrozoa may be said to date from the classical researches of Trembley, published in 1744, upon species of *Hydra*. He divided specimens into pieces of various sizes and shapes, and from various portions of the body, securing entire polyps from each.

He turned polyps inside out and had them live and thrive for months. He also grafted portions of one upon another with equal facility and success.

These researches were later repeated by Baker, who, while confirming some of Trembley's experiments, was not able to do so for all of them. Similar results were had by Rösels von Rosenhoff in 1755, who in addition claimed to have secured entire polyps from fragments of tentacles.

In 1878 Engelmann again repeated Trembley's experiments, and with results very similar to those of von Rosenhoff.

Marshall in 1882 was not able to secure successful grafting or eversion, but regeneration of polyps from portions of ten-

tacles, the body of the tentacle becoming the body of the polyp, and in turn forming new tentacles.

Nussbaum in 1887 and 1890 successfully everted specimens and described the process by which the body layers righted themselves. He secured no regeneration from detached tentacles.

Ischikawa in 1889 experimented upon eversion of Hydras successfully and described the process by which the layers readjusted themselves, differing in some respects from the account of Nussbaum.

In 1895 and 1898 Wetzel grafted specimens and secured union between portions of same species; but while bits of a different polarity united, the distinctness of polarity was only exceptionally modified. He secured only temporary unions of portions of different species.

Miss Peebles in 1897 undertook the determination of the smallest portions of Hydra capable of regeneration.

Rand in 1898 worked out interesting results on the "Regulation" of regeneration in Hydra.

Comparatively little has been done of an experimental nature upon Hydroids.

Loeb in 1891 carried on an extensive series of experiments on regeneration and heteromorphosis among Hydroids, chiefly of the genera *Antennularia*, *Eudendrium*, and *Tubularia*, with the object of determining as far as possible the external conditions and causes which affect life and growth. Light and gravitation were shown to have a profound influence in determining many of the phenomena.

In 1892 Miss Bickford conducted experiments on *Tubularia tenella*, confirming in many points the work of Loeb.

Davenport in 1894 conducted regenerative experiments upon *Obelia commisuralis*, with a view to determining the distribution of generative or embryonic tissue in various regions of the Hydroid.

The contribution of the present writer to the general subject in 1897 has already been cited.

Driesch in 1897 reviewed the work of Loeb and Bickford on *Tubularia*.

Summary and Conclusions.

1. Tubularian Hydroids readily react to experiments directed toward regeneration or grafting.
2. They exhibit in most cases striking illustrations of heteromorphosis.
3. They show no marked polarity, readily coalescing in either oral or aboral relations.
4. They show no sex differentiation of tissues limiting the process of grafting.
5. Closely allied species may be intergrafted.
6. Different genera have not been successfully grafted.
7. Hydromedusae respond to regenerative and grafting experiments with almost equal readiness.
8. No definite aboral grafting of Medusae has been successfully made.
9. Medusae show throughout a sharp polar orientation.
10. No heteromorphic results have been shown among Medusae.

SYRACUSE UNIVERSITY, June, 1899.

LITERATURE CITED.

1. 1896. BORN. Ueber Verwachsungsversuche mit Amphibienlarven. *Archiv f. Entwickl.* Bd. iv, p. 349.
2. 1897. CRAMPTON. Biological Lectures.
3. 1894. DAVENPORT. Regeneration in Obelia and its Bearing on Differentiation in the Germ-Plasma. *Anat. Anzeiger.* Bd. ix, p. 283.
4. 1897. DRIESCH. Studien über das Regulationsvermögen der Organismen. *Archiv f. Entwickl.* Bd. v, p. 389.
5. 1878. ENGELMANN. Ueber Trembley's Umkehrungsversuch an Hydra. *Zool. Anzeiger.* Jahrg. i, p. 77.
6. 1897. HARGITT. Recent Experiment on Regeneration. *Zoöl. Bull.* Vol. i, p. 27.
7. 1898. HARRISON. Growth and Regeneration of Tail of the Frog Larvae. *Archiv f. Entwickl.* Bd. viii, p. 430.

8. 1889. ISCHIKAWA. Trembley's Umkehrungsversuch an Hydra nach neuen Versuchen erklärt. *Zeit. f. wiss. Zool.* Bd. xlix, p. 433.
9. 1891. LOEB. Untersuch. z. physiologischen Morph. der Thiere. Würzburg.
10. 1896. LILLIE. On the Smallest Parts of Stentor Capable of Regeneration. *Journ. Morph.* Vol. xii, p. 239.
11. 1882. MARSHALL. Ueber einige Lebenserscheinungen des Genus Hydra. *Zeit. f. wiss. Zool.* Bd. xxxvii, p. 664.
12. 1887. NUSSBAUM. Ueber die Theilbarkeit der lebendigen Materie, etc. *Archiv f. Mikr. Anat.* Bd. xxix, p. 265.
13. 1890. NUSSBAUM. Die Umstülpung der Polypen. *Archiv f. Mikr. Anat.* Bd. xxxv.
14. 1891. NUSSBAUM. Mechanik der Trembleyschen Umstülpungsversuche. *Archiv f. Mikr. Anat.* Bd. xxxvii, p. 513.
15. 1899. RAND. Regeneration and Regulation in *Hydra viridis*. *Archiv f. Entwick.* Bd. viii, p. 1.
16. 1897. PEEBLES. Experimental Studies on Hydra. *Archiv f. Entwick.* Bd. v, p. 794.
17. 1744. TREMBLEY. Mem. pour servir a l'histoire d'un genre de Polypes, etc.
18. 1895. WETZEL. Transplantationsversuche mit Hydra. *Archiv f. Mikr. Anat.* Bd. xlv, p. 273.
19. 1898. WETZEL. Transplantationsversuche mit Hydra. *Archiv f. Mikr. Anat.* Bd. lii, p. 70.
20. 1896. LOEB. Ueber den Einfluss des Lichtes auf die Organbildung bei Thieren. *Archiv f. Physiol.* Bd. lxiii.
21. 1894. BICKFORD. Notes on Regeneration and Heteromorphosis of Tubularian Hydroids. *Journ. Morph.* Vol. ix.

BIBLIOGRAPHY AND PUBLICATION.

Zoölogical Bibliography and Publication. — Second Report of the Committee, consisting of Sir W. H. FLOWER (Chairman), Professor W. A. HERDMAN, Mr. W. E. HOYLE, Dr. P. L. SCLATER, Mr. ADAM SEDGWICK, Dr. D. SHARP, Mr. C. D. SHERBORN, Rev. T. R. R. STEBBING, Professor W. F. R. WELDON, and Mr. F. A. BATHER (Secretary).

The Committee wishes to state clearly that it has no wish, even if it had the authority, to lay down laws for zoölogists or for publishing bodies and editors. It is, however, plain that many are grateful for some guidance, and the Committee hopes that it may serve as a medium for conveying to those who need it the general opinion of the experienced. There are also difficulties which, though they appear to some insuperable, may possibly be surmounted in ways that have been communicated to the Committee.

(1) 'That each part of a serial publication should have the date of actual publication, as near as may be, printed on the wrapper, and, when possible, on the last sheet sent to press.'

Five correspondents do not see the use of this, thinking that the date on the wrapper is enough, and that in the case of annual publications the date of the year suffices. The Committee would point out that wrappers are constantly lost in binding, and that periodicals are often broken up by specialists or secondhand booksellers, the consequent loss of date causing much trouble to workers of a later day. To avoid this, the Cincinnati Society of Natural History would add the date at the head of each paper, while *Natural Science* prints the month and year across every page opening. Some societies, e.g., the Philadelphia Academy, issue a certificate of dates at the end of the volume. The Liverpool Biological Society 'put at the head of each paper the date when it is read, and are willing to add the date when it is printed off'; neither of these dates are necessary, and they may be misleading. In most cases the actual day of publication is immaterial, especially in cases where no new species are described, but at least the month should always be given, and the Committee does not see that there need be any difficulty in doing

this. If some unforeseen delay does occur, the date can always be rectified with a date stamp.

(2) 'That authors' separate copies should be issued with the original pagination and plate numbers clearly indicated on each page and plate, and with a reference to the original place of publication.'

The Committee believes this to be a most important recommendation, and its view is supported by all the zoölogists consulted. Nevertheless, many leading publications continue to issue authors' copies repaged, and often without reference to volume number, date, or even the name of the periodical. The remedy is so simple that the Committee urgently appeals for its universal application.

(3) 'That authors' separate copies should not be distributed privately before the paper has been published in the regular manner.'

It is a curious fact that on this question editors take a different line to working zoölogists. All the latter who have discussed the matter agree with the Committee as to the extreme inconvenience caused by the general custom. Among the editors, however, nine (*i.e.*, nearly one-quarter) protest against the present recommendation. The objectors represent small societies which publish at lengthy intervals, and their reasons are: that it is not fair to an author to prevent him from receiving his separate copies for perhaps a year; that it is not to the advantage of science that work should thus be delayed; that a society which did this would receive fewer contributions and lose its members. In brief, the argument is: 'We are too poor to publish properly; therefore we must allow authors to publish improperly.' This form of argument suggests an easy remedy, and one that, on the informal suggestion of the Committee, has already been put into practice by the Liverpool Biological Society and by the R. Physical Society of Edinburgh. The remedy is this:

In cases where a volume or part can only appear at long intervals, each author that requires separate copies of his paper for private distribution before its publication in the volume or part should be permitted them only on this condition — that, for every month before the probable issue of the volume, a certain number of copies — say five — should be placed by him in the hands of the society or its accredited publisher, in order that they may be offered for sale to the public at a fixed price. Further, that the society, for its part, should announce the publication, with price and

agent, of their papers to some recognized office, or to some such paper as the *Zoologischer Anzeiger*. The details of expense must be settled between the author and the society.

- (4) 'That it is desirable to express the subject of one's paper in its title, while keeping the title as concise as possible.'

It is satisfactory to find no objections raised to this recommendation, since there is no doubt that there is room for much improvement in this direction. Such phrases as 'Further contributions towards our knowledge of the . . .', or 'Einige Beobachtungen über . . .', or 'Essai d'une Monographie du genre . . .', might well be dispensed with as superfluous. The ornithologist who, in 1895, published a book with a title of ninety-one words would seem to have forgotten the functions of a preface.

On the other hand, it is pointed out that certain periodicals, such as the *Bulletin de la Société Entomologique de France* and the *Sitzungsberichte der Gesellschaft naturforschender Freunde zu Berlin*, publish communications without any title, to the constant confusion of naturalists. The Committee begs to urge the reform of this practice, in which it can see no advantage.

- (5) 'That new species should be properly diagnosed, and figured when possible.'

The only comment on this is the proposed omission of the words 'when possible.' With this the Committee sympathize, but wish to avoid all appearance of laying down a law that would constantly be broken.

- (6) 'That new names should not be proposed in irrelevant footnotes or anonymous paragraphs.'

Naturally nobody supports such actions as are here objected to, but since some have doubted the possibility of the latter, it is as well to state that the suggestion was based on an actual case occurring in the Report of a well-known International Congress. The proposal of a new name, without diagnosis, in a footnote to a student's text-book, or in a short review of a work by another author, is a by no means rare occurrence. The Committee believes that such practices are calculated to throw nomenclature into confusion rather than to advance science.

- (7) 'That references to previous publications should be made fully and correctly if possible, in accordance with one of the recognized sets of rules for quotation, such as that recently adopted by the French Zoölogical Society.'

Dr. Paul Mayer, of Naples, writes : ' Most authors are extremely idle in making good lists of literature themselves, and even oppose my correcting them according to our rules. There ought to be some training in this at our Universities.' This is confirmed by one or two other editors, but not all have the energy of Dr. Mayer. Some, indeed, oppose the word 'fully' on the ground that it leads to waste of time and space. The Committee would explain that the reference to a particular set of rules was intended merely as a guide to those who have not had the training that Dr. Mayer would like to see ; they would also point out, in the words of the editor of the Cincinnati Society of Natural History, that ' what may be intelligible to the specialist is very puzzling to the general student.' Nowadays, when so many zoölogists work with the aid of authors' separate copies, it is an enormous convenience to them to have the title of the paper at least indicated, and not merely the volume, date, and pagination given. The Committee, therefore, cannot agree that this suggestion involves a waste of time.

Communications with reference to this Report should be addressed to F. A. Bather, Natural History Museum, Cromwell Rd., London.

BIOLOGICAL BULLETIN.

THE EARLY STAGES IN THE DEVELOPMENT OF THE HYPOPHYSIS OF *AMIA CALVA*.

J. M. PRATHER.¹

THE results of my work on *Amia* do not agree with the common assumption that the pituitary body is always of epiblastic origin. This disagreement has led me to an examination of the literature on the subject to see if there is sufficient unity of opinion among recent investigators to warrant such a general conclusion. As a result it is found that a diversity of opinion still prevails, and that it is unsafe to predict its origin in any class of vertebrates.

A brief classification of the various views and their respective advocates is of interest in this connection: K. E. von Baer ('28), Huschke ('54), and F. Schmidt ('62) believed the hypophysis to be a modified part of the brain. Reichert ('40) and His ('68) claimed that it is derived from the end of the chorda. Reichert ('61) and Rathke ('61) believed it to be derived from the pia mater, each having changed his earlier view. Dursy ('68) maintained that it is of threefold origin — from the foregut, the chorda, and the brain.

The above represent the earlier but now generally discarded hypotheses. The more modern views, some of which were also held by the older anatomists, may be grouped as follows:

1. *That the hypophysis is of hypoblastic origin*, as held by

¹ This study was undertaken at the suggestion of Dr. A. C. Eycleshymer and completed under his direction in the Department of Anatomy and Histology in the University of Chicago during the summer of 1899.

Rathke ('38), Luschke ('60), Kölliker ('61), Miklucho-Maclay ('70), W. Müller ('71), His ('75), Hatschek ('81), Dohrn ('81), Owen ('82), Balfour and Parker ('82).

It should be emphasized that, in general, this was claimed for the particular form investigated, but not claimed to hold true for all vertebrates.

2. *That the hypophysis is of epiblastic origin*, as claimed by Goette ('72), Balfour ('74), Mihalkovics ('74), Kölliker ('76), Cattie ('81), Julin ('81), Dohrn ('82), Kraushaar ('83), Johnson and Sheldon ('86), Orr ('87), Scott ('87), Kupffer ('90), Lundborg ('94), Dean ('96), Haller ('96), Braem ('98), Minot ('98).

It should be added that the majority of these observers not only claimed the hypophysis to be of epiblastic origin in the particular form examined, but also believed this to hold good for the Vertebrata.

3. *That the hypophysis is partially of hypoblastic and partially of epiblastic origin*, as positively maintained by Kupffer ('93), Valenti ('95), and Nussbaum ('96), and considered probable by Hoffmann ('85), Orr ('87), and Gaupp ('93).

The researches made by the above-named writers show that the organ varies so much in its development and structure in the different forms that generalizations should be made with extreme caution, until more extended and precise observations have been made.

While its development has been more or less carefully traced in animals representing nearly every group of vertebrates, I find that the Ganoids have received but little attention. Kupffer has described and figured its earliest stages in *Acipenser sturio*. This author finds such an unusual mode of development that it seems possible that he has misinterpreted certain structures connected with the development of the sucking discs, for, as the sequel will show, at a certain stage in their formation in *Amia* these discs present appearances very similar to those regarded by him as the beginnings of the hypophysis. A fuller discussion is reserved for a later page.

Balfour and Parker have figured some of its early stages in *Lepidosteus osseus*, but have given no adequate description of its origin. Judging from their figures and few

remarks, its early history is in most respects quite similar to that in *Amia*.

Dean has figured a very early and a very late stage in its formation in *Amia calva*. No description is given of these stages, but the generalizations made therefrom seem, in the light of my researches, to be somewhat questionable, and to them I shall later recur.

Concerning the observations by Professor Minot I know nothing further than the simple statement in *Science*, Feb. 18, 1898, that he had confirmed and extended the results of B. Haller. As Haller's extensive observations did not embrace the Ganoids, I infer that this statement does not pertain to Minot's investigations upon *Amia*.

With these facts before us it will be seen that a detailed account of the development of the hypophysis in the last mentioned form will not be superfluous.

The sections for this study were placed at my disposal by Dr. Eycleshymer, and consist of series of sagittal, horizontal, and transverse sections of the embryo and larva, in many successive stages of development, up to and including the thirty-fifth day after fertilization. All ages given in the following descriptions are reckoned from the time of fertilization.

Many examinations were made with the oil-immersion and all the drawings have been made with the aid of the camera lucida. The literature consulted, and that to which reference has been made, consists for the most part of papers enumerated by Kupffer¹ and Haller.² Since a very complete bibliography is given by each of these authors it would be superfluous to duplicate them in the present paper.

DESCRIPTION OF STAGES.

The earliest stage in the formation of the hypophysis, clearly recognizable as such, is found in an embryo a few hours before

¹ Kupffer, C. von, "Die Entwicklung des Kopfes von *Acipenser Sturio* an Medianschnitten untersucht." München und Leipzig. 1893. "Die Deutung des Hirnanhanges," *Sitzungsber. d. Gesell. f. Morph. u. Phys. in München*. Jahrg. 1894.

² Haller, B., "Untersuchungen über die Hypophyse und die Infundibularorgane," *Morph. Jahrb.* 1897.

hatching, as shown in Figs. 2 and 3. In order to show the relations of position which the various organs bear to one another prior to the differentiation of the hypophysis, I figure a median sagittal section of an embryo surrounding about 245° of the circumference of the yolk, corresponding to an age of about 148 hours (Fig. 1). At this stage the foregut (*fg.*) is seen to have formed as far back as the posterior limit of the third primary vesicle (*hb.*), where the endoderm forming its wall is reflexed upon itself and passes forwards again over the surface of the yolk (*y.*). By tracing the foregut through the successive sections of the series, we find it to be a very broad cavity compressed dorso-ventrally until its upper and lower walls are in close apposition in the region immediately under the base of the first primary vesicle, which rests directly upon its dorsal wall. Its walls are slightly separated both anterior and posterior to this region. In the median plane, as shown by the figure, the cavity can be traced forwards to a point somewhat anterior to the front wall of the brain (*br.*). From its anterior end a diverticulum may be traced on either side in front of the brain, nearly to the dorsal median plane. These endodermal diverticula later become transformed into the larval adhesive organ, as has recently been shown. The endodermic layer increases in thickness anteriorly until a maximum thickness is attained in the adhesive organ just mentioned. The walls of the endoderm cells cannot be distinguished, owing to the great amount of yolk material found in them.

The ectoderm at the anterior end is invaginated in two places. The upper invagination (*o.*) is merely a depression within the adhesive organ which is developing beneath the ectoderm by diverticula from the foregut, as just described, pushing the ectoderm outwards in the form of a circular ridge (*ao.*) around the snout. The lower invagination is the involution for the stomodaeum (*st.*), and has already pushed inwards nearly to the endoderm surrounding the foregut. This invagination is on a lower plane than the foregut and is directed distinctly downwards, making a small angle with that plane.

The large cavity (*c.*) between the brain and the epiblast in front is for the most part filled by the developing adhesive

organ. But surrounding this may be seen mesodermal cells which form a strand running downwards between the stomodaeum and foregut and connecting with those forming the heart (*h.*) below. The chorda extends no further forwards than the posterior margin of the hind brain. The brain is not as clearly delimited from the ectoderm above as the figure indicates. The first primary vesicle (*fv.*) is evaginated at its base both in front and behind, giving rise to the recessus opticus (*ro.*) and the infundibulum (*in.*) respectively. The base of this vesicle is arched considerably and conforms closely to the dorsal wall of the foregut.

In an embryo about 160 hours old (Fig. 2) marked changes may be noted in all the organs described in the preceding stage. The adhesive organ has broken through the ectoderm, forming a semicircular row of sucking cups on either side of the snout. The section, not being exactly vertical, passes through one of these cups (*sc.*) on the dorsal side. The space between this organ and the brain is now filled with mesoblastic cells (*ms.*). The stomodaeal invagination has deepened a little, owing to the development of the sucking disc over and in front of it. By the vertical growth of the brain the anterior end of the alimentary canal has been pushed downwards to a level with the mouth fold, so that the ectoderm lining the stomodaeum is in close contact with the endoderm forming the wall of the foregut. The oral plate (*op.*) thus formed is on the point of breaking through, and the point of fusion of the endoderm roofing the gut with the ectodermal roof of the stomodaeum is scarcely recognizable. There is no fold of the ectoderm comparable to "Rathke's pocket" discernible, nor is there an endodermal fold comparable to "Seesel's pocket." The ectoderm immediately over the stomodaeum is much thickened and composed of large cells of irregular shape loosely aggregated.

Fig. 3, an enlarged portion of the same section, will show that this ectodermal layer (*ec.*) terminates rather abruptly at the point of junction (*pf.*) with the endoderm (*en.*), at which point the cells are seen suddenly to become smaller. While they are somewhat disconnected in this region, they soon become arranged into three definite layers running back under the ante-

rior part of the thalamencephalon as far as the posterior limit of the optic chiasma (*oc.*). Here, immediately under the central part of the thalamencephalon (*lpo.*), the hypoblastic cells may be seen to have assumed a different shape and size, and the number of layers to have increased. From roundish or ovoid cells they become long, spindle-shaped, much smaller, and arranged in crescent-shaped layers about six in number and fitting one within another, so that the whole mass of cells over a region about $180\ \mu$ in length is nested in this peculiar manner. This is the first trace of the hypophysis (*hy.*) recognizable. Back of this differentiation the cells of the endoderm are again of the same rounded or ovoid shape as in front and arranged in two layers. Thus, I repeat, there can be detected no fold or overgrowth of ectoderm to give rise to the hypophysis nor evagination of the endoderm. Its cells are differentiated *in situ* apparently by longitudinal division of the cells constituting the roof of the foregut.

The chorda at this stage extends forwards under the hind brain to near the tip of the infundibular fold, which has greatly enlarged. The base of the thalamencephalon has elongated and is now a single layer of cells (*lpo.*) posterior to the chiasma, resting closely upon the dorsal wall of the foregut.

In a larva early in the eighth day, shortly after hatching, a sagittal section (Fig. 4) shows the oral plate broken in the center. But sections of the series on either side show the membrane or its remnants still intact, stretching across the oral cavity from a point near the tip of the now forming lower jaw to a point (*op.*) on the dorsal side of the cavity just forwards of the anterior limit of the thalamencephalon where it rests upon the dorsal wall of the foregut. It is thus seen that the position of the membrane relative to surrounding parts has not changed from the preceding stage. The floor of the foregut, however, has dropped down in the anterior part, making a rather deep cavity (*fg.*) immediately below the thalamencephalon and behind the oral plate. The dimensions of the thalamencephalon have increased vertically and decreased antero-posteriorly. The infundibular fold has apparently widened, but not deepened. The optic chiasma has greatly thickened, while the base of the

brain remains one cell in thickness and rests flatly upon the dorsal wall of the gut and the hypophysis. The sucking disc (*sc.*) is relatively at its largest size. The cavity between it and the brain is filled with a denser aggregation of mesoblastic cells than in the previous stage.

Fig. 5 shows the base of the brain and the hypophysis of the same section more highly magnified. The hypophysis now measures $150\ \mu$ in length and $28\ \mu$ in thickness. This apparent shortening may be due to the uncertainty of the limits of the organ in the previous stage, or to individual variations in the two larvae. That peculiar nesting of the cells in the hypophysis, as described above, will be seen to continue, but the cells are now perceptibly larger, while the epithelial cells (*em.*) forming the roof of the mouth beneath the hypophysis have again attained the size and shape of those with which they are continuous in front and behind. The basal layers of endoderm, however, still seem to be dividing, adding new layers to the base of the hypophysis by proliferation. The roof of the mouth is now three layers deep both before and behind the hypophysis.

A transverse section (Fig. 6) of a larva about eight days old, through the posterior part of the hypophysis and the infundibulum, shows that the organ is at this stage convex on the dorsal side in a transverse plane, fitting into a concavity in the base of the infundibulum lying closely upon it. An examination of the successive sections forwards and backwards from this shows that the upward convexity gradually diminishes forwards, but more abruptly backwards. This figure, in connection with the figures of longitudinal sections of larvae both older and younger, shows that the organ is now approximately lenticular in shape. In transverse, as in longitudinal sections, that characteristic nesting of the cells differentiated to form the hypophysis is found to prevail, a basin-shaped stratum of lenticular cells formed from the deeper layers of the endoderm, with other similar strata, each successively smaller, fitting into the previous ones, until we get a nest of seven or eight basins, while the hollow of the upper basin is filled to a rounded fullness with cells of a more nearly uniform diameter, rounded or

polygonal, and with no evident order of arrangement. This fact is strikingly evident, that the limits of the organ, both longitudinally and transversely, mark very accurately the borders of the area over which the brain is in such close contact with the hypoblast.

The mouth cavity posterior to the oral plate is now rather shallow but very wide. Just over the oral epithelium roofing the mouth, on either side of the hypophysis and the infundibulum, the internal carotid arteries (*v.*) are forming, while dorsal to these, in the folds formed between the infundibular lobe and the thalamocoele above, may be seen two other large cavities (*v'*), the cavities of the preoral somites.

A sagittal section of a larva about nine days old (Fig. 7) shows the base of the thalamencephalon (*fv.*) to be a single layer of columnar cells where it rests on the hypophysis, but thickened in front for the chiasma and likewise behind in the infundibular region, which has grown larger. The end of the chorda approaches very near to the infundibulum, but remains separated therefrom by mesoblastic cells, which may be seen to grow in between the brain and mouth roof to the limits of the hypophysis at either end. The lower jaw is relatively further back, its tip now lying under the anterior end of the hypophysis. The epithelium of the mouth is continuous beneath the hypophysis, but the basal layer of endoderm appears to continue to divide, adding new cells to the hypophysis by proliferation. The marked difference between the shape and arrangement of the cells in the upper and lower portions of the organ may be noticed still. At the posterior end the body is distinctly separated from the mother layer, while at the anterior end its cells take on more and more the character of the mother layer as we go forwards. The organ measures in this plane $196\ \mu$ in length by $41\ \mu$ in thickness.

A transverse section of a slightly older stage (Fig. 8), during the tenth day, shows an oval-shaped organ $130\ \mu$ in breadth by $64\ \mu$ in thickness. It is clearly separated from the endoderm at either side, so that it seems to be wholly differentiated, but still lying in a depression in the endoderm caused by the transformation of the cells of this layer into cells of the hypophysis.

The characteristic stratification on the ventral side and the irregular grouping of the cells above are still pronounced. Very nearly in the center, between these lower strata and the cells above, is a nearly spherical cavity (*l.*) about $16\ \mu$ in diameter, the first lumen that has been recognized. This lumen is not a longitudinal slit, as it is found only in this section; and since it is enveloped by no distinct membrane, and the cells have no definite arrangement about it, it has the appearance of an intercellular space. The mesoderm (*ms.*) is seen to grow between the brain and the mouth up to the sides of the hypophysis, just as at the ends. It would thus appear to be a biconvex body fitting into concavities in the brain above and the endoderm below.

In a larva ten days old, of which Fig. 9 is a sagittal section of the hypophysis and parts adjacent and Fig. 10 a transverse section more highly magnified, the organ has become entirely separated from the mouth roof, which has again become two cells in thickness beneath it. A strand of mesoderm (*ms.*) passes between the hypophysis and mouth roof and is continuous at the sides with the thickened mesoderm differentiating to form the cranial cartilages. An outfold of the infundibular wall at its posterior lower margin, $33\ \mu$ by $39\ \mu$ internal measurement, is the first stage in the formation of the infundibular gland, or saccus vasculosus (*sv.*). The single layer of cells characterizing the base of the infundibulum and the base of the thalamencephalon is continued into the saccus, but here the cells are columnar, with nuclei at their outer ends, in marked contrast with the cells in the brain wall contiguous. The chorda (*ch.*) runs forwards under the hind brain almost in contact with the roof of the mouth, and its cephalic end (*chh.*) abuts against this infundibular process with a strand of mesoderm between. This mesoderm grows into the space about the hypophysis, and a thin strand of it runs between the organ and the mouth roof below.

The hypophysis now measures in longitudinal section $143\ \mu$ by $57\ \mu$, in transverse $136\ \mu$ by $66\ \mu$. A lumen may be seen in the center,—an oval cavity with a membrane surrounding. About this lumen the spindle-shaped cells seem to be arranged in a radiate fashion, with long axes pointing towards it.

In a larva about fourteen days old a median sagittal section (Fig. 11) shows marked changes in surrounding parts, with but little change in the hypophysis. The saccus vasculosus has grown until it measures at this stage 120μ in length, and from 6μ in width at its point of origin to 27μ at its widest part. Its tip now touches the base of the hind brain. The columnar cells which characterized it in the preceding stage are yet very noticeable, the transition to the rounder cells of the infundibular wall above being very abrupt, while the transition to the cells of the infundibular base is more gradual. Beneath, the hypophysis rests directly upon a strand of fibrous tissue (*ms.*) continuous before and behind with the perichondrium surrounding the sphenoidal cartilages, which are encroaching from all sides. The membrane roofing the mouth (*em.*) has become widely separated from the hypophysis, and in it dental protuberances and glandular cells have differentiated. The hypophysis has now attained a size of 156μ by 56μ , and has several small spherical or lenticular cavities which do not communicate.

A sagittal section of the hypophysis of a larva about the same age is shown in Fig. 12 strongly magnified. It will be seen that a few mesodermal cells have come to lie between the hypophysis and the brain, forming a thin layer separating the two organs. With this interpolation of mesoblastic tissue the lobing of the hypophysis begins, and this continues to increase with age. A large central oval lumen is conspicuous at every stage, while in the section here figured, several smaller lumina are met with as one examines the sections of the series. The principal lumen in this case measures 19μ by 6μ , and can be detected in but two sections, showing that its shape is lenticular. The smaller ones are more nearly spherical, and each is found in but a single section. No communication between them can be found. The hypophysis in this larva measures 164μ in length by 57μ in thickness. The characteristic radiate arrangement of the cells about the lumina is very noticeable.

Fig. 13 shows the relations of the hypophysis to surrounding parts, at a stage about one day older, in a transverse section through the middle of the organ. The oral epithelium has

further differentiated. The lateral cartilages (*sk.*) of the skull are closely encroaching from the sides, enclosing the internal carotids (*bv*). These cartilages are connected by a strand of the perichondrial membrane which runs beneath the hypophysis. The organ is almost perfectly lenticular in cross-section, measuring $180\ \mu$ by $70\ \mu$. A principal lumen is seen in the center, but others are found at other positions in sections in front and back of this one.

In a larva between twenty-two and twenty-six days old, a cross-section (Fig. 14) shows the hypophysis appreciably enlarged, measuring now $220\ \mu$ by $75\ \mu$. An irregular but distinct lobing may be seen, more pronounced on the upper than on the lower side. In this way, from a very symmetrical organ in the fifteen-days stage, we get here a marked asymmetry. It is here firmly adherent to the infundibulum, but quite apart from the wall of the cranial cavity. This separation from the base of the cranial cavity may be considered an artificial condition, as succeeding stages invariably show it to be in contact with this wall. The distinct upward bend in the infundibular base, the enlargement and gradual encroachment of the lateral cartilages, and the advancing differentiation of organs in the oral epithelium may be remarked.

The same section of the hypophysis highly magnified (Fig. 15) shows its histological structure to present some interesting features. The large central lumen with its very definite lining membrane is a striking object. It is ovoid, measuring $24\ \mu$ by $14\ \mu$. Other spheroidal but smaller lumina are to be found near the tip of the different lobes as they are traced by sections, but communication of these lumina with each other, or with the central lumen, cannot be definitely proved. It seems that communication may be had through very narrow channels representing connected intercellular spaces between rather definite rows of cells and running from the lumina of the lobes towards the central lumen. But apparently these channels are closed before reaching the lumen. Such spaces may be seen in the figure (*sl.*) running out into the lobes to the right and to the left. It will be noticed that the cells maintain a rather definite and orderly arrangement about the central lumen. There is a

row encircling the lumen in a radiate manner, long spindle-shaped in the upper and lateral parts, more rounded below. The other cells appear to have a general tendency to arrange themselves in rows pointing towards the center of the organ, but this arrangement is modified by a secondary tendency to be grouped about the secondary lumina and the longitudinal channels. A marked feature of this and succeeding stages is the indistinctness or total obscurity of the nuclei.

A sagittal section at this stage shows the same peculiar arrangement of cells about the lumina and channels in the separate lobes. And the other characters of the hypophysis are similar to those shown in the cross-section. The anterior sphenoidal cartilage has now advanced to the posterior part of the chiasma nearing the hypophysis, the posterior to a point not far from the posterior point of the saccus, while dense skeletogenous tissue continues to and below the saccus nearly to the hypophysis. The perichondrium, as before, stretches across from one cartilage to the other below the organ. The basilar artery is now well formed, running along the base of the hind brain up into the fold between the hind brain and the primary forebrain. A blood vessel may be seen also just posterior to the hypophysis beneath the point of origin of the saccus. The saccus has enlarged, and in its cavity may be seen abundant granular secretions.

Passing from this stage to a stage between thirty and thirty-five days, a sagittal section (Fig. 16) shows all parts much enlarged. The finger-shaped saccus measures internally $311\ \mu$ by $18\ \mu$ at its narrow opening into the infundibulum, and $77\ \mu$ at its widest part. The granular secretions noticed in the previous stage have increased in amount. While the sphenoidal cartilages have enlarged and strengthened compared with the condition in the previous stage, they have approached very little nearer to the hypophysis; but the connective tissue between the hypophysis and the roof of the mouth has considerably increased. The hypophysis at this stage has attained a size of $359\ \mu$ by $96\ \mu$. Increased lobing is not apparent from the figure, but the series shows a great increase in the number of lobes and of the lumina in them. The arrangement and char-

acteristics of the cells are not enough different from those described in the preceding stage to call for special remark.

A comparison of sagittal with transverse sections (Fig. 17) demonstrates that in shape the organ retains the general lenticular form acquired early in its formation. It measures now 284μ in breadth by 88μ in thickness. The lateral cartilages (*sk.*) have advanced far in towards the hypophysis, so that it may be clearly seen that the organ lies in a distinctive space surrounded by cartilages on all sides—the pituitary fossa. Small bits of connective tissue may be seen between the hypophysis and the brain in the folds of the former, and also in the recesses between the lobes on the under side. The infundibular base is folded more or less in conformity to the lobing of the hypophysis. No evident communication between the cavities nor ducts opening to the exterior have been observed at this, the latest stage studied.

A horizontal section of the hypophysis (Fig. 18) of a larva about 20 mm. in length, thirty days old, shows that the lobing of the organ is principally around the edge; that its general shape is circular in this plane, lenticular as a solid; that a cavity may be found near the end of each well-formed lobe, which may possibly communicate with the central lumen by a very narrow indistinctly defined channel; that the cells are, in general, arranged in a double row around each lobe, the space between the rows constituting the channel mentioned; that the cells are arranged radially about the lumina. The section figured is not exactly in a horizontal plane, but dips a little posteriorly and to the right, so that the lobes mostly appear to be on the anterior side, but are in reality of approximately the same number in each half. The organ is here seen to be enclosed in the sella turcica, which is far advanced in its formation. The infundibulum fits closely upon its upper surface, the projections of the one fitting roughly into the depressions of the other. No blood vessels can at this stage be seen entering the organ, nor nervous tissue be found connecting it with the brain. It seems not to have become glandular as yet.

SUMMARY.

In distinction from an epiblastic origin, as found in most forms, my observations lead me to believe that the hypophysis is of hypoblastic origin in *Amia*.

Prior to the differentiation of the hypophysis the foregut extends far forwards; by diverticula the hypoblast reaches even the dorsal side of the head in front of the brain. At this time the stomodaeal involution is below the front end of the foregut. The diverticula later sever their connection with the foregut and are metamorphosed into the larval adhesive organ. The hypoblast unites with the epiblast on the last day before hatching, seventh day, forming the oral plate at a point forwards of the anterior limit of the brain.

There is no indication of an overgrowth of epiblast between the brain and the foregut, nor is there an invagination from the stomodaeum to give rise to the pituitary body. Neither does an outfold from the hypoblast occur to form it. Its first stage is found near the close of the embryonic period, about 160 hours, as a local differentiation of hypoblastic cells in the dorsal wall of the mesenteron, immediately under the thalamencephalon where the base of the brain is in close contact with the hypoblast. The intimate fusion of the base of the first primary vesicle with the hypoblast, from a time long before the stomodaeum has united with the foregut until after the hypophysis has become well differentiated, seems to me to preclude the possibility of any epiblastic tissue entering into its composition, and leads me to think that its origin is probably due to a mechanical cause. This region of fusion is far back of the oral plate, which remains intact for several hours after the differentiation of the hypophysis has begun.

The growth of the hypophysis is at first apparently due more to the enlargement of the cells first differentiated to form it than to the addition or multiplication of cells. It appears to remain in genetic connection with the mother layer until about the tenth day, when it becomes wholly delimited therefrom by an ingrowth of mesoblastic tissue between it and the mouth roof.

During this early period there is a distinct stratification in the arrangement of cells in its lower portion, as if formed in successive strata by proliferation from the mother layer. This stratification is not apparent in the upper part of the organ, and is no longer seen in the lower part after the ninth day.

The first lumen is formed about the ninth day, from which time on an increasing number of lumina is to be found as the organ develops. A lumen appears near the end of each lobe. These lumina are oval or spherical cavities, and definite channels of communication have not been observed, though indications that such channels are forming are found in the arrangement of the cells about the longitudinal axes of the lobes. The cells have a tendency to arrange themselves radially about the lumina.

The organ is almost perfectly lens-shaped until about the fifteenth day, when the formation of lobes begins with the interpolation of mesoblastic cells between the hypophysis and the brain. The number of lobes multiplies from this time on till the thirty-fifth day, beyond which stage observations have not been made. The lobes form chiefly around the periphery of the lenticular body.

The organ is thus, at this stage, a spongy body with isolated cavities, rather than a complex of glandular tubules which so frequently characterize it. The nuclei of the cells become indistinct or wholly disappear by the twenty-second day. This may indicate a glandular modification, but no evident glandular secretions have been detected. No duct nor external opening of cavities has been observed.

No arteries or blood vessels are to be found in it at the latest stage examined. Neither have nerve fibers been seen connecting it with the brain.

The cranial cartilages developing from the mesoblast increase in size and strength from about the tenth day, until at the thirty-fifth day they closely surround the organ on all sides, forming the pituitary fossa, in which the organ lies in close contact with the infundibulum on the dorsal side, but separated from the mouth by a fibrous strand of connective tissue on the ventral.

The saccus vasculosus begins to form about the tenth day and steadily enlarges, until at the thirty-fifth day it forms a process of the infundibulum in the shape of a glove-finger, extending directly backwards under the base of the medulla and parallel with the base of the cranium.

This is very nearly its position in the adult brain as figured by Allis.¹ At no stage, therefore, is it in close association with the hypophysis, which connection is found to be true in so many cases. Abundant granular secretions are found in it at the twenty-second-day stage, and these increase in amount thereafter.

GENERAL REMARKS.

The course of development of the hypophysis in *Amia* as described in the preceding pages, when compared with the descriptions given by other observers, both in *Amia* and other Ganoids, will be seen to present some striking peculiarities.

Dean² says: "The hypophysis is by no means as important an element in the development of the head in *Amia* as in other Ganoids. Its appearance is late and inconspicuous. It has not been found in stages earlier than that of Fig. O (*hatching time*),³ and even here its presence is not definite. At the most the position of its lumen can be recognized as the line *HY*, formed by the arrangement of cells immediately below the region of the recessus opticus. These cells are apparently ectodermal, for they are arranged in a continuous line with the cells of the formative epiblast of the dorsal wall of the stomodaeum, but, on the other hand, their ventral limit cannot be distinguished from the entodermal cells roofing the foregut."

In what respect the development of the hypophysis is less important in *Amia* than in other Ganoids is not clear. In point of size and position its relations are almost exactly the same as in the other Ganoids, so far as the larval stages have been examined. As for its appearance being "late and incon-

¹ Allis, Edward Phelps, "The Cranial Muscles and Cranial and First Spinal Nerves in *Amia Calva*," *Journ. of Morph.* Vol. xii, No. 3, Pl. XXXVIII. 1897.

³ Italics are mine.

² "On the Larval Development of *Amia Calva*," *Zool. Jahrb.*, p. 667. 1896.

spicuous," I may say that it arises, as will be shown, at a time intermediate between the time of its first occurrence in the other two forms in which this point has been determined, and it is certainly a conspicuous, I might say, striking differentiation of cells, even in its fundamentals, although it may not be a prominent object in point of size or in the distinctness with which its limits may be fixed. The actual time of its appearance I have found to be several hours earlier than the time assigned by Dr. Dean, as a comparison of Fig. 4 with his Fig. O plainly proves. His figure and description indicate a stage nearly the same as my Fig. 4, which is shortly after the time of hatching. Although in his figure the oral plate is still unbroken, the cavity of the foregut posterior to it has noticeably deepened by the dropping downwards of its basal wall, while in the stage I figure the cavity is of about the same depth, and the oral plate is only severed at its middle point, remaining intact at the sides. While in Fig. 4 the cells, which he would call the beginning of the hypophysis, may be said to be "continuous with the cells of the formative epiblast," being in the same plane with them, there is no other evidence of a connection and certainly no ground whatever for considering them derived therefrom, as the enlargement of this section (Fig. 5) makes very clear their differentiation from cells of the hypoblast. But its earliest stage is found (Figs. 2 and 3) some hours previous to this stage, far back of the oral plate and the epiblast, as a well-defined modification of cells of the basal layer of hypoblast where the post-optical lamina of the brain rests closely upon it. Considering the common assumption of embryologists that its origin is from the epiblast, my observations become of interest, since I believe that I have proved beyond question that the hypophysis is of hypoblastic origin, and I do not at present understand how Dean could have considered it otherwise.

Balfour and Parker¹ state in a footnote: "We have not been able to work out the early development of the hypophysis as satisfactorily as we could have wished. . . . Were it not for

¹ "On the Structure and Development of *Lepidosteus*," *Trans. Roy. Soc. Pt. ii*, p. 379. 1882.

the evidence in other types of its being derived from the epiblast, we should be inclined to regard it as hypoblastic in origin." They speak of it as an invagination of the oral epithelium, without stating whether this invagination is anterior or posterior to the oral plate. Presumably from the previous statement they consider it derived from the roof of the foregut posterior to the oral plate. They figure a transverse section of the anterior part of the head of an embryo on the ninth day after impregnation, showing an invagination from the mouth roof with a thickened, solid, conical process extending upwards into the cranial cavity. This, with the exception of the invagination, is very similar to its condition in cross-sections of *Amia* late in the seventh day, passing through the anterior end of the organ, as shown in Fig. 6. They also figure transverse sections through the anterior and posterior ends of the hypophysis of an embryo eleven days old, where in front it is still in connection with the oral epithelium, while behind it is constricted from that layer. These sections indicate relations almost exactly the same as transections of the organ in *Amia* shown in sagittal section (Fig. 7) at an age of not quite nine days. Comparing these few sections of *Lepidosteus* and the few words of description with the conditions which I find in *Amia*, it seems that the hypophysis undergoes a nearly parallel series of changes in the two forms from the time of its primary origin to a late larval stage, but that corresponding embryonic stages are met with in *Amia* from one and a half to three days earlier than in *Lepidosteus*, and the corresponding larval stages are found earlier and earlier as the animal advances in age. This justifies, so far as the hypophysis is concerned, Dean's statement that "the organogeny of *Amia* progresses more rapidly than in *Lepidosteus*."

I find no lumen earlier than the ninth day, when it appears as a single nearly spherical cavity near the center of the organ, rather than a longitudinal slit, as indicated by Dean in Fig. O. Furthermore, as my sections show, the lumen is never a longitudinal slit, even at the late stage of thirty-five days, corresponding to Dean's Fig. Q of a four-weeks-old larva which shows it as such. Instead of a single lumen at this late period,

I find several lumina which apparently do not communicate. Neither do my observations agree with those of Balfour and Parker on *Lepidosteus* at a corresponding age, where the hypophysis is described as "small, not divided into lobes, and provided with a very small lumen"; whereas in *Amia* at this period it is much lobed and possesses numerous spherical lumina.

Kupffer (*loc. cit.* p. 59) has described a very early and unusual formation of the hypophysis in *Acipenser sturio*. According to him the organ arises during the second day after fertilization by an invagination of the basal layer of the ectoderm on the dorsal side of the head, in front of the brain, between the yet unclosed neuropore and the sucking disc. This invagination grows downwards and backwards until it comes in contact with the endoderm, and then, before the close of the second day (forty-five hours), by a rupture of this layer, it communicates with the alimentary canal, which communication persists for about twenty-four hours. But before the stomodaeum has formed a connection with the foregut, this union of the hypophysis with the latter has broken off (*i.e.*, by the sixty-fourth hour), and the lower blind end of the tube becomes swollen into a hollow bulb. This bulb gradually enlarges and migrates backwards to a position between the dorsal wall of the gut and the base of the thalamencephalon, while the remaining part of the tube, namely, the stalk, gradually atrophies and has wholly disappeared by the time the embryo is hatched (eighty-seven hours).

Haller has expressed a doubt as to Kupffer's interpretations. This doubt is based upon the indistinctness of the parts observed at this early time. From Kupffer's own words it would seem that the cells were greatly obscured by food-yolk. I give the passage quoted by Haller: "Die Entodermzellen sind noch mit Dotter überladen, die Zellen der Epidermis und des Hirnes schon fast dotterfrei, aber diejenigen Ektodermzellen, die in der Bildung der gleich zu besprechenden Organe (*Hypophy-senanlage*, etc., Haller) eingehen, zeigen noch denselben Dotter-vorrath wie die Elemente des Entoderms und sind daher durch Färbungen von diesen nicht zu unterscheiden. Die Abgren-

zung der einzelnen Theile dieser Region gelang mir erst unter vergleichender Prüfung nahe auf einander folgender älterer Stadien."

I likewise believe that Kupffer has misinterpreted what he saw, but for an entirely different reason from that given by Haller, yet based upon the same passage.

Miss Phelps¹ has recently shown that the "larval adhesive organ" in *Amia* develops at about the time that Kupffer assigns to the formation of the hypophysis in *Acipenser*. The adhesive organ begins as a diverticulum from the endoderm anterior to the brain, which later becomes divided into a pair of diverticula whose connection with the endoderm becomes broken off after a time. *Acipenser* and *Lepidosteus* each possesses a sucking disc, or adhesive organ, similar to that in *Amia* and at a corresponding developmental period. It would therefore seem that they should justly be regarded as homologous organs, and, further, we should naturally expect them to have a similar ontogeny. Previous to the observations of Miss Phelps this organ has been assumed to be of ectodermic origin (Dean for *Amia*, Balfour and Parker for *Lepidosteus*, and Kupffer for *Acipenser*).

In my own studies on *Amia*, as described on a preceding page, I observed these diverticula connecting the foregut with lateral masses of cells lying on either side of the snout and between the anterior end of the brain and the overlying epidermis, filling nearly the whole of this pre-cerebral region. These masses of cells present an appearance exactly like the cells walling the foregut, being laden with much yolk just as Kupffer describes, and could not be distinguished from them. The narrow channels can be traced from the cavity of the foregut upwards through the masses to near the epidermis on the dorsal side where they end blindly. The cells of the epidermis and brain contiguous to these present a very different appearance, due to their freedom from yolk.

I was at a loss to account for these canals connecting the enteric cavity with organs on the dorsal side of the head, until

¹ "On the Development of the Larval Adhesive Organ in *Amia*." Abstract in *Science*. March 10, 1899.

I saw the account of the development of the adhesive organ by Miss Phelps. I at once concluded that I had not only confirmed her discovery, but had also found wherein Kupffer has probably erred in his interpretation of the structures which he believed to be connected with the development of the hypophysis, but which to my mind have an entirely different meaning. The statement by Kupffer, that the cells overladen with yolk going to form the hypophysis are not to be distinguished from the elements of the endoderm, receives its explanation in the fact that they are endodermal cells growing out from the foregut by evagination. The canal which, according to Kupffer, connects the foregut with the dorsal ectoderm is nothing else, in the opinion of the writer, than this median diverticulum from the foregut, or possibly one of the lateral diverticula resulting from it, running up through the adhesive organ to the point of closure of the neuropore. A slightly oblique dorso-ventral section through this region in *Amia* during the sixth day of development would show strikingly similar appearances to those figured and described by Kupffer in *Acipenser*.

As a phylogenetic interpretation of the singular development of the hypophysis in *Acipenser*, Kupffer holds that we here have traces of the ancestral mouth which opened above the sucking disc and in front of the brain. Its origin from the stomodaeal roof as found in most vertebrates, he claims, is a secondary condition due to a migration downwards from its original position, compelled by the great development of the fore brain in these forms and the concomitant degeneration of the adhesive discs, whose remnants, he thinks, are still to be found in the fold between "Rathke's pocket" and the oral plate.

This explanation fails to account for its origin in *Amia* and *Lepidosteus*, in which the sucking disc is still large and functional, and whose fore brain is little different from that found in *Acipenser*; and yet in them the hypophysis arises at a point far back of the sucking disc and under the brain.

It may be considered highly probable that the hypophysis is

endodermic in origin in *Lepidosteus* as well as in *Amia*, and I venture the prediction that a further study of *Acipenser* will demonstrate the same for that form. The foregut extends far forwards in each of the Ganoids in which its development has been studied, and the writer believes that in all these it arises from the endoderm. A study of the figures of the head parts in those animals in which the hypophysis is undoubtedly of ectodermal origin shows that in them the foregut stops short of the infundibulum, while in some, at least, of those forms in which its origin is questionable — ectodermic or endodermic — the foregut and stomodaeum meet at an intermediate point, directly under the base of the thalamencephalon. These facts lead me to suggest that mechanical factors, acting at the point of fusion of the brain base to the oral roof, may play an important part in determining its development from this or that layer.

If my observations be confirmed, that the hypophysis in *Amia* is derived from the hypoblast, will this fact strengthen Kupffer's hypothesis that it represents a degenerated "paleostome," or will it rather revive the old theory of Dohrn, that it represents a portion of a canal connecting the alimentary tract with an exterior dorsal opening through the thalamencephalon and the epiphysis and accordingly believed to be homologous with the invertebrate pharynx? I find nothing in its structure or its relations, other than its point of origin, which can be interpreted as evidence in support of either hypothesis. I have given what I believe to be the facts, but must leave their interpretation to the maturer judgment that will come from the more complete and extended observations of the future.

HULL ANATOMICAL LABORATORY,
September 22, 1899.

ABBREVIATIONS.

<i>A.</i>	anterior.	<i>m.</i>	mouth cavity.
<i>ao.</i>	outfold of ectoderm caused by the developing adhesive organ.	<i>mb.</i>	mesencephalon.
<i>br.</i>	wall of brain.	<i>ms.</i>	mesoderm.
<i>bv.</i>	blood vessels.	<i>o.</i>	involution of ectoderm between the sucking discs.
<i>c.</i>	cavity occupied by adhesive organ in its young stages.	<i>oc.</i>	optic chiasma.
<i>ch.</i>	chorda.	<i>op.</i>	oral plate.
<i>chk.</i>	cephalic end of the chorda.	<i>ov.</i>	optic vesicle.
<i>ec.</i>	ectoderm.	<i>P.</i>	posterior.
<i>egm.</i>	egg membrane.	<i>pf.</i>	point of fusion of ectoderm with endoderm.
<i>em.</i>	epithelium of mouth.	<i>ro.</i>	recessus opticus.
<i>en.</i>	endoderm.	<i>s.</i>	strand of fibrous tissue continuous with the perichondrium.
<i>ep.</i>	epiphysis.	<i>sc.</i>	sucking cup.
<i>fg.</i>	foregut.	<i>sk.</i>	skeletal cartilages.
<i>fv.</i>	first primary vesicle of brain.	<i>sl.</i>	cleft between rows of cells.
<i>h.</i>	position of heart.	<i>st.</i>	stomodaeum.
<i>hb.</i>	third primary vesicle of brain.	<i>sv.</i>	saccus vasculosus or infundibular gland.
<i>hy.</i>	hypophysis.	<i>v.</i>	internal carotid artery.
<i>in.</i>	infundibulum.	<i>v'.</i>	head cavities.
<i>l.</i>	lumen of hypophysis.	<i>y.</i>	yolk.
<i>lpo.</i>	lamina postoptica.		
<i>ls.</i>	crystalline lens.		

EXPLANATION OF FIGURES.

FIG. 1. Median sagittal section of the anterior portion of an embryo surrounding about 245° of the egg's circumference. About 148 hours. $\times 60$.

FIG. 2. Median sagittal section of the anterior portion of an embryo a few hours before hatching time. About 160 hours. $\times 60$.

FIG. 3. The base of the thalamencephalon, the foregut, and the stomodaeum of the same section under stronger magnification. $\times 190$.

FIG. 4. Median sagittal section through the anterior portion of a larva soon after hatching, early in the eighth day. $\times 60$.

FIG. 5. The roof of the mouth and the hypophysis of the same section more highly magnified. $\times 190$.

FIG. 6. Transverse section through the brain and hypophysis of a larva about eight days old. $\times 60$.

FIG. 7. Median sagittal section through the hypophysis and base of the thalamencephalon of a larva nearly nine days old. $\times 190$.

FIG. 8. Transverse section through the hypophysis and base of the infundibulum of a larva during the tenth day. $\times 190$.

FIG. 9. Median sagittal section through the hypophysis and infundibulum of a larva ten days old. $\times 60$.

FIG. 10. Transverse section of the hypophysis and base of the infundibulum of a larva ten days old. $\times 190$.

FIG. 11. Median sagittal section through the hypophysis and infundibulum of a larva fourteen days old. $\times 60$.

FIG. 12. Median sagittal section through the hypophysis and adjacent parts of a larva about the same age as the preceding. $\times 190$.

FIG. 13. Transverse section of the hypophysis and infundibulum of a larva about fifteen days old. $\times 60$.

FIG. 14. Transverse section through the hypophysis and infundibulum of a larva between twenty-two and twenty-six days old. $\times 60$.

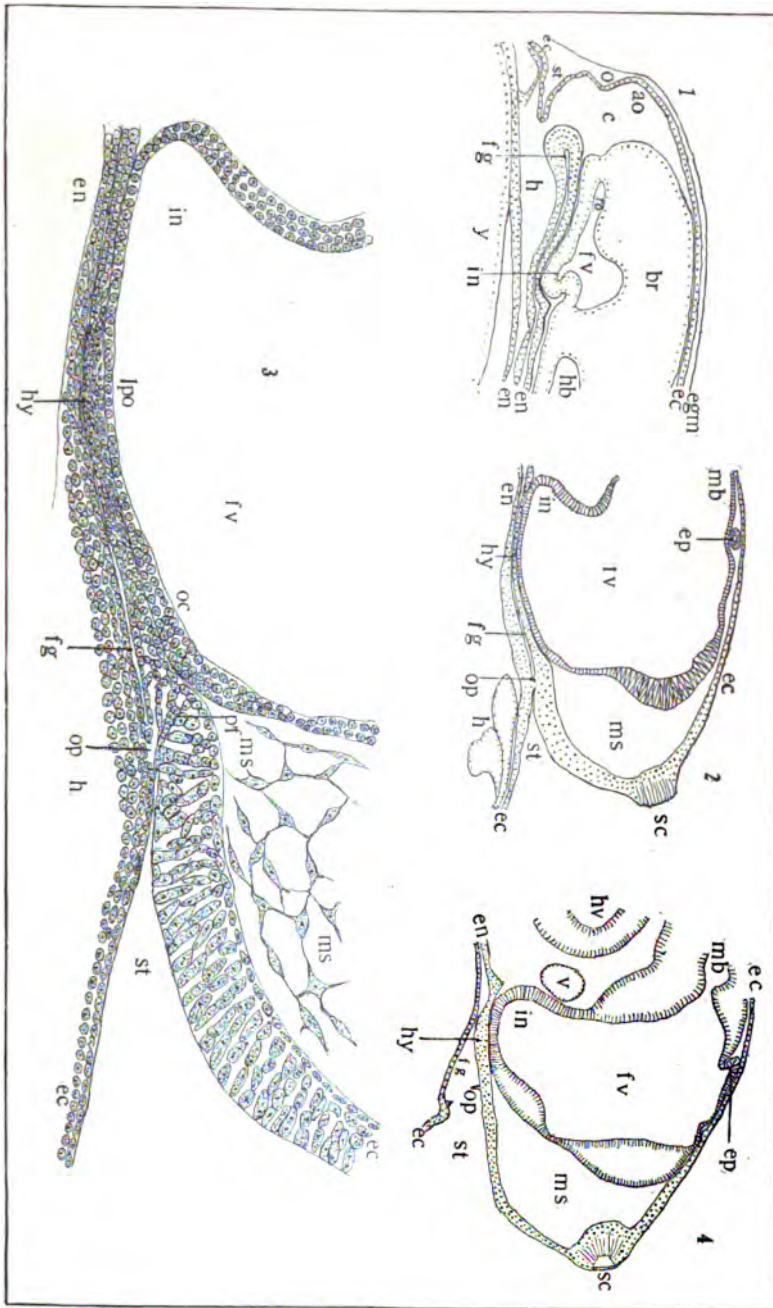
FIG. 15. The hypophysis of the same section more highly magnified. $\times 215$.

FIG. 16. Median sagittal section through the hypophysis and infundibulum of a larva between thirty and thirty-five days old. $\times 60$.

FIG. 17. Transverse section through the hypophysis and infundibulum of a larva of the same age as the preceding. $\times 60$.

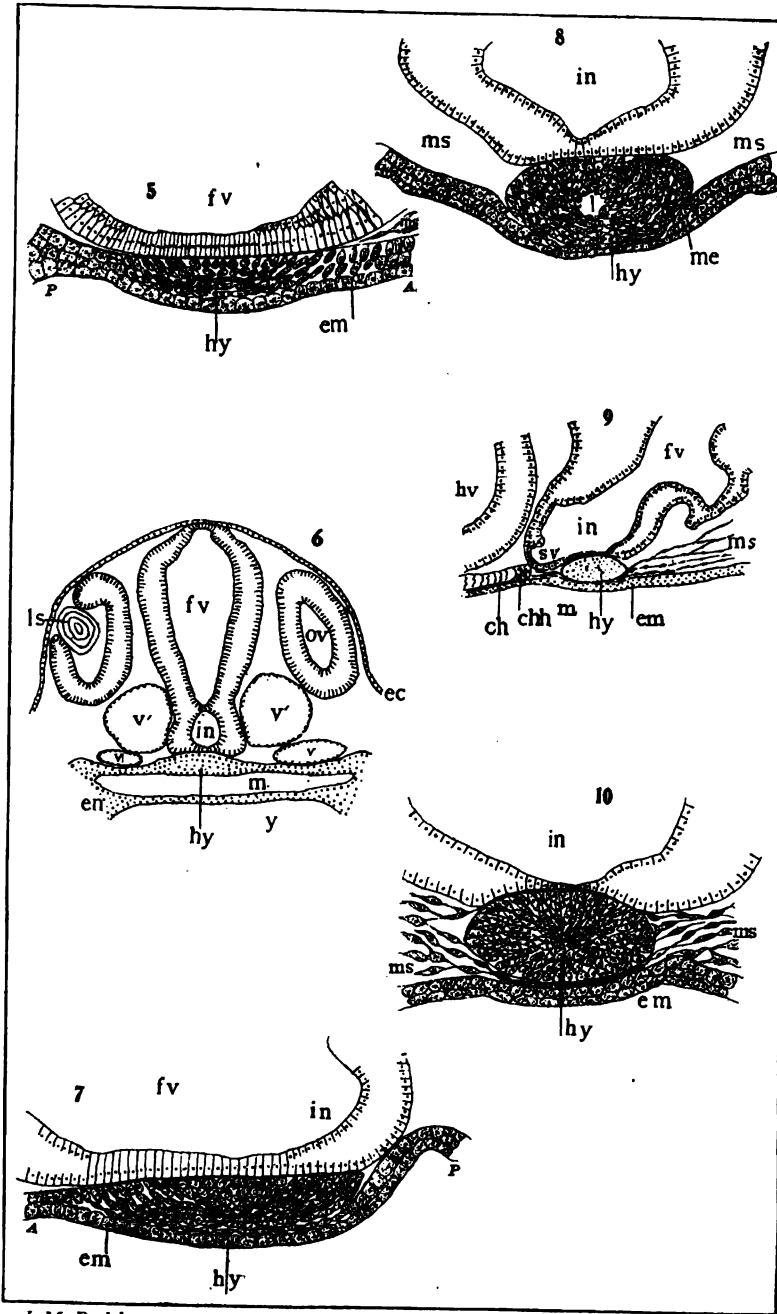
FIG. 18. A nearly horizontal section through the hypophysis of a larva about thirty days old. $\times 105$.

Plate I.



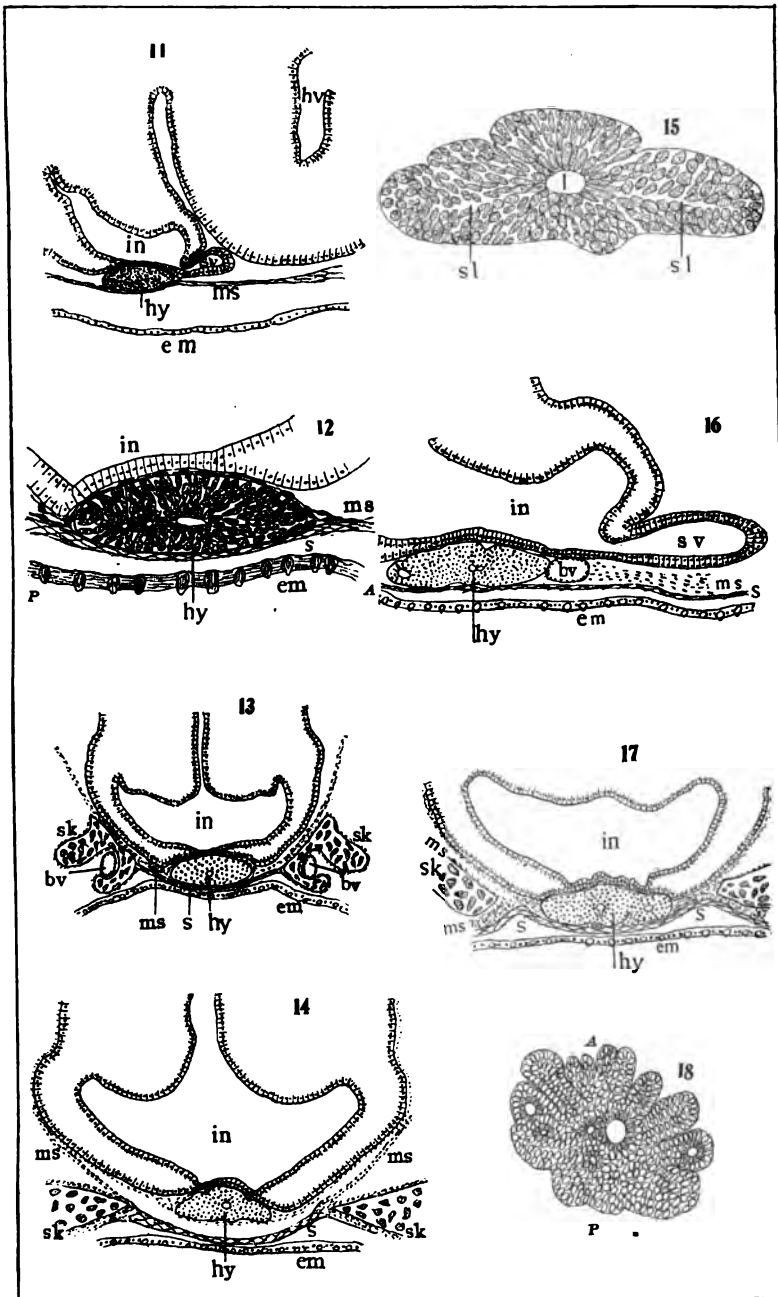
J. M. P. del.

Plate II.



J. M. P. del.

Plate III.



J. M. P. del.

AN EXTRAORDINARY NEW MARITIME FLY.

VERNON L. KELLOGG.

THERE have been established recently two new¹ families of flies, to which I have to add a third. In the case of two of these three new families the members show great divergence from the usual dipterous condition. The three genera of Wandolleck's new family, Stethopathidae, are wingless and are without halteres. The thorax is greatly reduced and the compound eyes are feebly developed. The mouth-parts are of the general sort possessed by the Nematocera, *i.e.*, a short lip-like labium without pseudo-tracheae, a distinct labrum, and a hypopharynx, but no mandibles nor maxillar lobes. Coquillet's new family, the Stenoxenidae, established for a single female fly, presents no such extraordinary characters as the Stethopathidae. "The shape and structure of the head, body, and legs, and the unusual development of the first antennal joint appear to indicate its nearest approach to the genus *Ceratopogon* of the family Chironomidae; but the venation as well as the general appearance of the insect is very different from anything now located in that family" (Coquillet).

There has come into my hands a number of specimens, 153 in all, of a fly which must prove of unusual interest to zoölogists and entomologists, both because of its peculiar habitat and of its extraordinary structural condition. This new fly can certainly not be ascribed to any known dipterous family; its affinities can only be determined in the most general way. I feel constrained to establish for it a new family, which may be called the Eretmopteridae.

The 153 specimens of the new form, 139 males and 13 females, and 1 female pupa, were collected on Dec. 27, 1898,

¹ Wandolleck, Bruno, "Die Stethopathidae, eine neue Dipteren-Familie," *Zool. Jahrb.* Bd. xi, pp. 412-441, Pls. XXV and XXVI. 1898.

Coquillet, D. W., "A New Dipterous Family Related to the Chironomidae," *Ent. News.* Vol. x, pp. 60 and 61 (figure). March, 1899.

by Mr. J. C. Brown, a student assistant in my laboratory, at Point Lobos, a rocky point on the Pacific Coast near Monterey, California. The flies, of which there were many ("thousands," says Mr. Brown), were resting or running on the surface of the ocean water of tide pools and had a tendency to gather in large numbers in "patches" and in "ball-like masses" on the water. None were seen below the surface, nor were any seen flying. They moved about on the surface of the water very rapidly. In the last week of March, 1899, I visited Point Lobos and searched carefully for the fly, examining the same tide pools on which

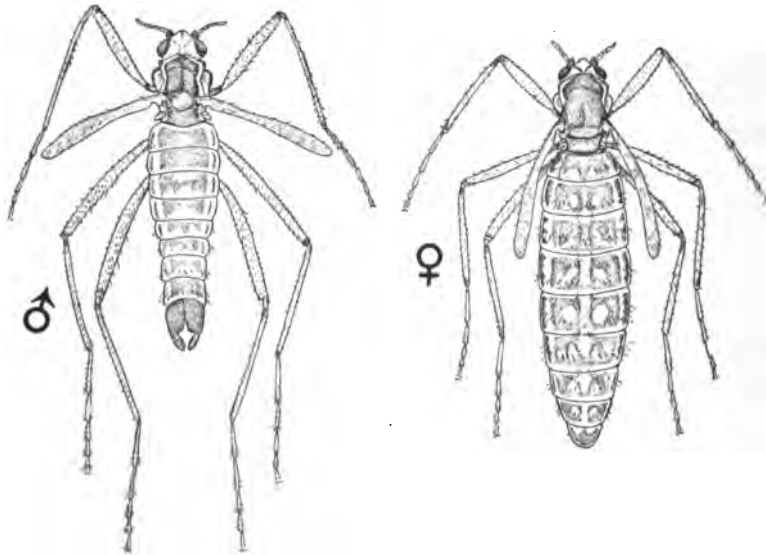


FIG. 1. — *Eretmoptera browni*; male and female.

Mr. Brown found his specimens; but no flies were to be found, nor were there any dipterous larvae or pupae in these pools. Mr. Brown also searched again in July and August without finding more specimens. So we have as yet no knowledge of the eggs and larvae, nor of the course of the life history of the fly.

The new form may be named and described as follows:

Eretmoptera browni nov. gen. et sp. — Male (Fig. 1). Length 2 mm. Head slightly broader than thorax; eyes widely separated, very small, very convex, hairy, and with rather large

facets; ocelli absent; antennae (Fig. 3, *ant.*) short, length 3 mm., 6-segmented, the basal segment wide and globose, the sixth segment longest, the second next, the third and fifth about equal, the fourth shortest, with a few short strong hairs on each segment, and the surface everywhere with a fine stiff pubescence. The mouth-parts are of simple nematoceros type, short, and with distinct labrum-epipharynx, maxillae, hypopharynx, and labium, mandibles absent; labrum-epipharynx (Fig. 2,

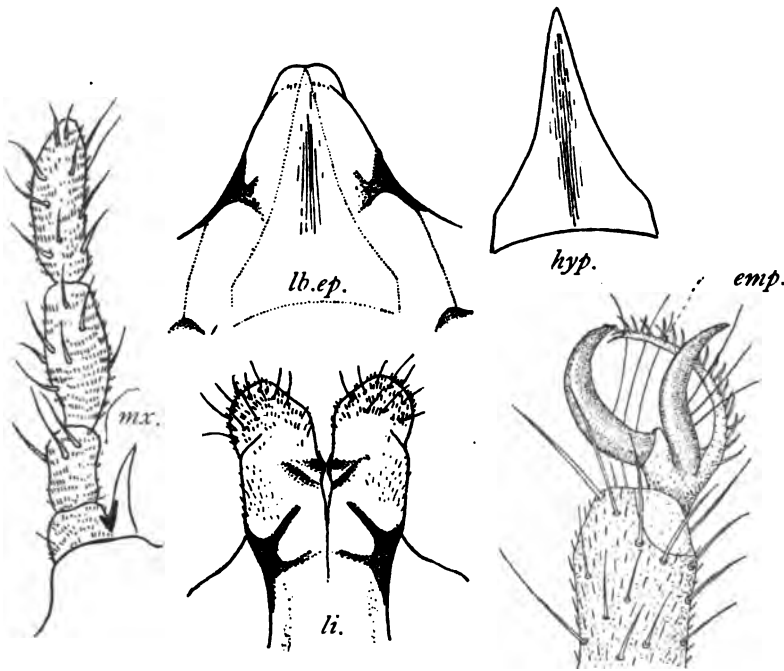


FIG. 2.—*Eretmoptera browni*: *mx.*, maxilla of male; *lb.ep.*, labrum-epipharynx of male; *li.*, labium of male; *hyp.*, hypopharynx of male; *emp.*, empodium of male.

lb.ep.) short, broadly triangular, with obtusely rounded tip; maxillae (Fig. 2, *mx.*) with short, weak, tapering, pointed lobe, and 4-segmented palpi, 3 mm. long; the palpi with last two segments longest and equal, and all the segments provided like the antennae with a few short stray hairs and a fine stiff pubescence; hypopharynx (Fig. 2, *hyp.*) elongate, triangular, as long as the labrum-epipharynx, but narrower and more acute; labium (Fig. 2, *li.*) short, lip-like, with free paraglossae, without pseudo-

tracheae. The face is whitish, with a median longitudinal dark line, and the antennary fossae with dark margins; the basal segment of the antenna is rather dark, the other segments pale. Thorax without bristles, dark above, pale beneath. Legs long and slender, whitish with blackish joints; middle and hind legs longest and equal, front legs only a little shorter; average measurement of middle leg, femur 1 mm., tibia 1 mm., tarsus 1 mm.; tarsus 5-segmented, segment 1 as long as segments 2, 3, and 4 together; segment 5 slightly longer than segment 4; tibiae of all legs with single apical spur; tarsal claws strongly curved, thickened at base, and with a few (three?) delicate spines on basal half; no pulvilli; empodium (Fig. 2, *emp.*) rather long, curving, filiform, and plumose or pectinate for its whole length. Wings narrow, strap-like, extending only to fourth abdominal segment, length .75 mm., and wholly without veins; whitish, somewhat wrinkled, and finely spinulose. These strange veinless wings are not specially thin or delicate, but, on the contrary, are rather thickened, the costal margin being especially thickened and perhaps folded. The halteres (Fig. 3, *h.*), or the structures which occupy the usual position of halteres, are not of the usual pedicel and knob type common among Diptera, but are minute, lobe-, or scale-like processes, appearing like rudiments of metathoracic wings; like the mesothoracic wings, they are rather thickened and are finely spinulose; they are widest at base and taper to a rounded tip; they average .08 mm. in length. Abdomen of nine segments, tapering gradually posteriorly; mottled gray and blackish above, lighter below, palest laterally; a few scattered, small, wholly inconspicuous hairs, the body appearing glabrous; external genitalia consisting of a pair of large, conspicuous, strong, articulated claspers (Fig. 3, *cl.*), which are covered with a pubescence.

Female (Fig. 1). — Length 2.5 mm., thus being one-fourth longer than the male; this extra length is all in the abdomen, which is markedly larger in every way than the abdomen of the male. The head and thorax are narrower than the robust abdomen, which is sub-cylindrical, tapering only slightly posteriorly. Eyes as in male very small, very widely separated, and hairy. Antennae (Fig. 3, *ant.*) only 4-segmented. Mouth-parts

essentially as in male, with, however, appreciable differences in shape; the labrum-epipharynx is narrower at base, and is more pointed apically; the labium with paraglossae separated farther back and slightly narrower. The reduced wings and halteres like those of male, the wings, length .85 mm., slightly longer. The abdomen consists of nine segments mottled blackish, with conspicuous white sutural spaces, caused by the distention of the abdomen. The external genitalia are inconspicuous. There is a short emarginate dorsal plate with rounded tips and a pair of small lateral processes. There appears to be no extrusible ovipositor.

Pupa of Female.—Among the many specimens collected by Mr. Brown, I find a single female pupa. This specimen throws the only light upon the condition of the immature life of the fly that we yet have. The pupa is of that simple unprotected, unmodified type characteristic of those flies, like the Cecidomyiidae and Mycetophilidae, whose pupae are protected by lying enclosed in plant tissue. There are no projecting breathing tubes like those of the aquatic pupae, and it would seem that the pupa was quite unfit for an aquatic life. And yet Mr. Brown took this pupa with the imagines from the surface of a tide pool. There is a puzzle here. The pupa may be described as follows: Length 2.5 mm. (as large as adult female). Immediately recognizable as pupa of the female from the similarity in size, shape, and markings. Abdomen just as in adult both as regards size, shape, color, and markings. The antennae, legs, and wings are folded on the lateral and ventral aspects of the anterior part of the body and extend backwards only to (hardly reaching) the posterior margin of the second abdominal segment. There are no external tracheal gills or elongated spiracles (breathing tubes). There are no bristles nor special clinging organs. The pupa is of a very simple, unmodified, unprotected type.

The "extraordinary" features of the external structure of the fly are the condition of the wings and halteres. The condition of the antennae and the empodium is also unusual. The reduction of the wings and loss of flight are accompanied by a reduction of the halteres, the flight-directing (?) organs. The

halteres being not wholly obsolete, but existing still in rudimentary condition, are especially suggestive in their likeness to rudimentary wings. The general affinities of the fly are shown by the character of the mouth-parts (and pupa) to be with the simpler nematocerous families. The habitat is unusual, but of course not unique. The discovery of the conditions of life of the immature stages may, however, give the matter of habi-

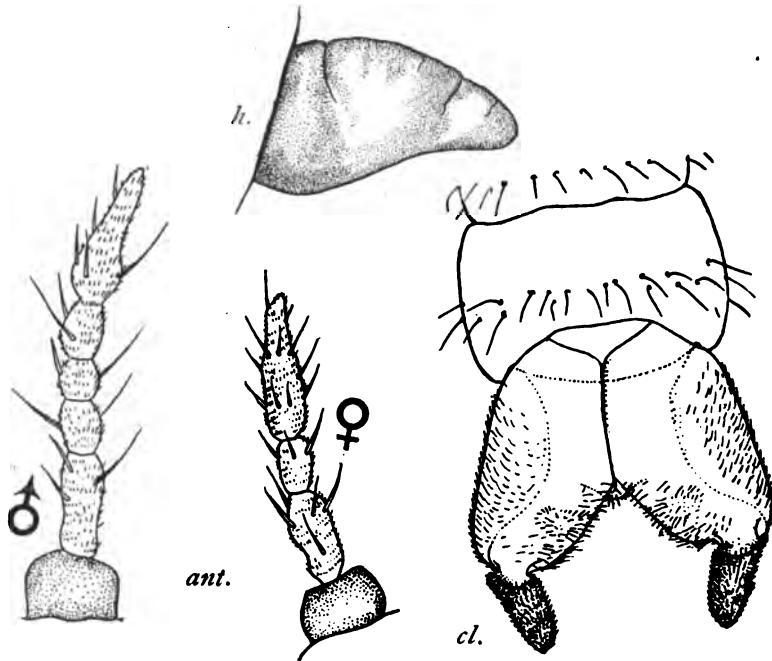


FIG. 3. — *Eretmoptera browni*: *ant.*, antennae of male and female; *h.*, balancer of male; *cl.*, claspers of male.

tat a very great interest. The fact that the imagines are unable to fly is to be remembered in connection with their presence on the tide pools. Are the thickened strap-like reduced wings used in locomotion at all?

Other tide-pool flies are known. Various winged forms are common at the verge of the water and must become accustomed to occasional watery overwhelmings. Wheeler¹ has

¹ Wheeler, W. M., "A Genus of Maritime Dolichopodidae New to America," *Proc. Cal. Acad. Sci.*, 3d series. Vol. i, pp. 145-152, Pl. IV. 1897.

described three species of Dolichopodidae which he found "flitting about in the spray of the breakers among the seaweeds on the rocks below high-water mark." Eaton, and later Verrall,¹ describe certain flies from the Kerguelen Islands which live on the verge of the tide. One of these forms, *Halirytus amphibius*, assigned to the Chironomidae, has 6-segmented antennae and rudimentary wings "reaching to the apex of the first abdominal segment." But it has but 2-segmented palpi, and the halteres are of the usual form. It was found "walking upon the surface of puddles and tide pools." And many of the flies were undoubtedly occasionally submerged at high tide, although none was seen under water.

STANFORD UNIVERSITY, CALIFORNIA.

¹ Verrall, in *Phil. Trans. Royal Soc.* Vol. clxxxvi, p. 247, Pl. XIV, Fig. 6. London, 1879.

ON THE VARIATION IN THE POSITION OF THE STOLON IN AUTOLYTUS.

P. CALVIN MENSCH.

IN the investigation of the variation in the position of the region of stolonization in bud-forming syllidians, observations were made upon four of the most abundant forms occurring along the Atlantic coast.

Three of these forms — *Autolytus cornutus*, *Autolytus varians*, and *Proceraea ornata* — may be found in abundance at Woods Holl in the summer months, during which time also the phenomenon of budding is most active. The fourth, *Proceraea tardigrada*, occurs in almost equal abundance at Beaufort, N. C. Of these *A. cornutus*, *P. ornata*, and *P. tardigrada* are solitary stolon-bearing, and invariably cast off the first stolon before a trace of a second stolon appears, while *A. varians* belongs to the chain-forming variety and may bear as many as eight stolons in various stages of development attached to the adult individual or so-called parent stock.

The greatest number of variations were observed in the chain-bearing form *A. varians*. The range in the position of the chain stolon in this species is from segment 19 to 58. The largest percentage of individuals were found to bear the chain on or between segments 30 and 38; a smaller percentage on or between segments 39 and 48, and an equal number between segments 25 and 29; fewer between segments 19 and 24, and a very few between segments 50 and 58.

In 155 individuals examined the following results were tabulated. (The upper numerals indicate the number of the segment to which the chain of stolons is attached; the lower, the number of individuals.)

A	{	Seg.	30	31	32	33	34	35	36	37	38
		Ind.	16	5	19	4	20	10	5	14	13

<i>B</i>	{ Seg.	25	26	29	39	40	41	45	48
	{ Ind.	5	4	9	9	6	4	2	3

<i>C</i>	{ Seg.	19	21	24	52	58
	{ Ind.	1	1	2	2	1

From this it will be noted that the position of the chain occurs most frequently on some segment in Table *A*, varying between segments 30 and 38, with a decided preponderance in favor of segments 34, 32, and 30, respectively. In other words, the greatest number of individuals have parent stocks of medium length, with from 30 to 38 segments; and the position of the chain on parent stocks of fewer or more numerous segments than this range occurs less frequently as the number of segments becomes less or greater.

Within the range in which the chain has been found to occur most frequently, it will also be noticed that several segments (31, 33, 36) bear the chain much less often than others, so that the chain-bearing phenomenon would appear to be confined more particularly to certain ones of the segments in this range (30, 32, 34, 37, 38).

In observing the sex of the individuals tabulated, it was noted that by far the greater majority of the specimens with the chain attached to the thirtieth segment or anterior thereto bore female stolons, while those with a great number of segments invariably bore male stolons. In this lot of specimens examined no individuals with female stolons were found with a parent stock of more than 41 segments, while those of 19, 21, and 24 segments all bore female stolons. On the other hand, those of 45 and more segments all bore male chains. Bearing in mind that the first stolon of the series in the chain is formed by the separation of a number of segments from the posterior part of the parent stock, this condition might possibly be due to the fact that the female stolons always consist of a far greater number of segments than do the male, and hence in the process of forming the first stolon leave a more reduced parent stock; while in the process of forming the first male stolon fewer segments would be required, and hence a longer parent stock left back.

In *P. ornata* and *P. tardigrada* variations in the position of

the stolon are of comparatively infrequent occurrence. The position of the stolon is very constantly on the thirteenth segment. In more than 200 specimens of *P. ornata* examined, not more than nine were found in which the stolon was attached to any other than the thirteenth segment; of these, six were found to bear the stolon on the fourteenth segment, while in three it was borne on the twelfth segment. In a single instance I have found the stolon as far forward as the eleventh segment. In all cases the variation was among individuals which bore male stolons.

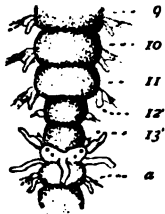
P. tardigrada shows even less variation than *P. ornata*. In 55 individuals examined, not a single case of variation was observed. Out of 110 individuals examined, Andrews (*Proc. U. S. Nat. Mus.*, Vol. XIV) obtained three specimens with the bud attached to the fourteenth segment.

Variations in the position of the stolon in the two species of *Proceraea* would accordingly be confined almost exclusively to occurrences of the position of the stolon either immediately anterior or posterior to a fixed segment, the posterior position being the most frequent form of variation.

In *A. cornutus* variations in the position of the bud are of still less frequent occurrence. Out of 178 specimens examined at a time when the forms were most abundant, not a single case of variation was observed. I have, however, found specimens in which the stolon was attached to the twelfth segment, but, as compared with *Proceraea*, such occurrences are very rare.

A further proof of the more constant occurrence of the bud on the thirteenth segment in this species is the fact that in individuals in which mutilation or severance of segments anterior to the region of budding has occurred, in the subsequent regeneration of new segments, the bud will not, as I have observed in *Proceraea*, develop its head from the tissues of the most anterior regenerated segment, but will first of all regenerate the lost segments of the parent stock, and following these produce the bud. The accompanying figure represents a specimen in which all the segments of the parent stock posterior to the eleventh were amputated and have been replaced by a series of new segments. The bud, instead of taking its origin in the

plane of new tissue formation, *i.e.*, on the twelfth segment, has followed the law of budding in this species and developed its head from the tissues of the fourteenth segment, and thus added two segments to the parent stock. The addition of new tissue to the parent stock, after amputation of one or two of its posterior segments, I have never been able to find in *Proceraea*, and so far as I have been able to carry my observations, I am convinced that such a condition does not take place, but that



9, 10, 11, old segments of parent stock; 12, 13, regenerated segments of parent stock; a, stolon with developing head.

instead, wherever new segments are formed following amputation, the head takes its origin from the tissues of the first new segment formed.

From these observations it would appear, therefore, that by far the greatest variation occurs in the chain-bearing form of *Autolytus*. The great range in the position of the chain, and hence the amount of variation, is modified by a condition to which I have already referred in a previous paper (*Journal of Morphology*, Vol. XVI, No. 2). I have there shown that

in this species, in the region in which new segments are being formed (region of proliferation), not all the newly formed segments are pushed back for the formation of new stolons, but that occasionally some of these segments become segments of the parent stock, and thus considerably increase its length. In this way I endeavor to account for the great length (40–58 segments) of the parent stock in the stouter and more developed specimens—a length which I have shown does not exist in the younger and more slender forms. This being the case, the true range of variation would have to be sought in such specimens only which are in the act of forming a first stolon by the separation of the posterior segments of the parent stock. Of such individuals I have found specimens sufficient to give a range from segment 19 to as high as segment 38. Thus while variation here, as identical with the mode of variation in the species investigated, does not present so wide a range as indicated in the tabulation, nevertheless it shows a far greater range and is of much more frequent occurrence in chain-forming

than in the solitary stolon forms. In marked contrast to the variations in *A. varians* is the constancy in the position of the bud in *A. cornutus*, where new segments even are formed for the maintenance of a fixed position of budding.

URSINUS COLLEGE, COLLEGEVILLE, PA.,

Nov. 16, 1899.

GORDIACEA FROM THE COPE COLLECTION.

THOS. H. MONTGOMERY, JR.

AT his death Prof. Edward D. Cope left to the Philadelphia Academy of Natural Sciences, among other alcoholic collections, a few specimens of Gordiacea. Among them is a new species, while the others are interesting from the standpoint of geographical distribution.

1. *Gordius aquaticus* (Linn). One ♂ from Haines Falls, Catskills, New York, U. S. A. This specimen is a typical one of this species, and is particularly interesting as coming from such an eastern locality of the United States; previous specimens I have described only from Mexico and California, while all others of the species seen by me from the eastern part of the continent belonged to the following subspecies:

2. *G. aquaticus robustus* (Leidy). Three ♂♂ from Austin, Texas, the first record from this State.

3. *Paragordius varius* (Leidy). One ♂ from the same locality.

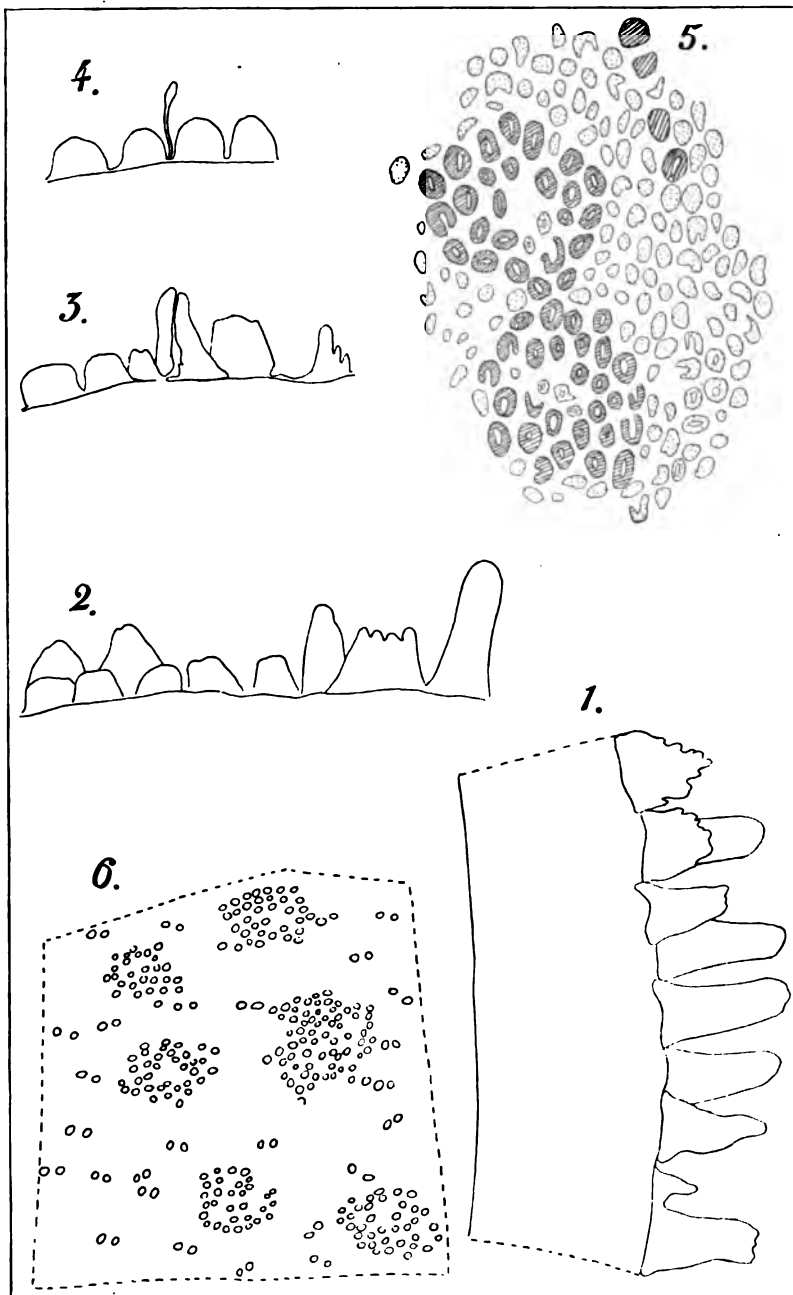
4. *Chordodes occidentalis* (Montg.). One ♂ from Texas, taken from the abdomen of a large grasshopper.

5. *C. Cameranonis*, n. sp. One ♂ from the West Coast, "Mazatlan or Panama." This type is in the collection of the Philadelphia Academy of Natural Sciences.

Form. — Body dorso-ventrally flattened, with slight median grooves. Anterior third of the body gradually tapering to a point, but the extreme end of the head truncated. Posterior end also attenuated, but less so than head; rounded terminally. A post-cloacal ventral groove is in the males of most species of the genus.

Cuticle. — Three main kinds of cuticular processes may be distinguished:

(1) The most abundant are low tubercles, which, on surface view (Fig. 5), appear more or less rounded or oval in outline, less frequently notched at one side, or sickle-shaped. While on



Figs. 1-4, portions of transverse sections of the cuticle, the cuticular processes shown only in outline; in Fig. 1 the outline of the fibrous cuticle is shown. (Zeiss homog. 1 mm. γ , oc. 2.) Fig. 5, surface view of a portion of the cuticle, showing both tubercles and papillae (the latter darker). (Oc. 4, obj. C.) Fig. 6, surface view of the cuticle, seen in water, to show only the arrangement of the groups of papillae, the tubercles not being shown. (Obj. A, oc. 4.)

surface view they appear to be more or less clearly separated from one another, on cross-section they are seen to be connected at their bases. On section (Figs. 2-4) most of them appear of a conical or rounded conical outline, with rounded or flattened smooth summits; but in some the summits are notched or toothed, and this is especially the case with those tubercles found on the margins of the papillar groups next to be described.

(2) Papillae, which on surface view of the cuticle (Fig. 5) appear darker than the tubercles just described, owing to their greater height. On the surface of the cuticle they are arranged in groups of two kinds (Fig. 6, where only these groups of papillae are shown, and none of the tubercles; and Fig. 5, where both tubercles and papillae are shown): (a) In larger groups consisting of from about twenty-five to fifty papillae (usually about forty) each; and (b) in pairs, the pairs being much more numerous than the larger groups. The line joining the two papillae of a pair lies in the transverse axis of the body, and not infrequently two or three pairs of papillae may lie in such close juxtaposition as to form transverse rows of four or six papillae each. In the larger groups of papillae the center of each group is occupied by papillae of less height than those on the periphery, or is devoid of papillae. These papillae may be readily recognized on surface view, in addition to their dark coloring, by the clearer central portion, which is often narrow and slit-like (Fig. 5).

Transverse sections of the cuticle (Figs. 1-4) show these papillae in two forms. First, there are long, comparatively slender, finger-shaped papillae, with smooth outlines and rounded summits; some of these attain a height nearly equal to the transverse diameter of the underlying fibrous portion of the cuticle. And, secondly, there are papillae of greater diameter at the base, but less height, which lie among the former kind, and may be distinguished by the irregular notching and tuberculation of their summits and sides. The clear central portion of these papillae seen on surface view is shown on cross-section to be a clear axial core running the whole length of the papilla and representing possibly a pore canal.

On none of these papillae have I been able to find terminal

hairs, such as are so frequently found in the larger papillae of many species of the genus; no hairs were to be seen on the cuticle on surface view examined in water and in balsam, nor yet on cross-sections examined with the one-twelfth immersion lens of Zeiss.

(3) Hyaline, club-shaped, delicate processes scattered sparingly over the cuticle (Fig. 4).

Color. — Black, head rufous-brown.

Dimensions. — Length, 425 mm.; greatest diameter, 2 mm. The structure of the genital organs showed this specimen to be sexually mature.

Comparisons. — This appears to be a well-marked species. In the arrangement of the papillae it bears some resemblance to *C. festae* Camerano, but differs from the latter in having no fine hairs ("corti e fini prolungamenti trasparenti") on the summits of the papillae and also in having no large, transparent, recurved hooks on the surface of the cuticle (*cf.* Camerano, "Monografia dei Gordii," *Accad. Real. Sci. di Torino*, 1897, p. 386, Taf. III, Fig. 38).

It is a pleasure to me to name this species in honor of Prof. Lorenzo Camerano, of Turin, who has given such an able systematic monograph of the group.

BIOLOGICAL SCHOOL,
UNIVERSITY OF PENNSYLVANIA, PHILADELPHIA,
December 11, 1899.

A PRELIMINARY ACCOUNT OF THE SPERMATO-
GENESIS OF *BATRACHOSEPS ATTENUATUS*,
POLYMORPHOUS SPERMATOGONIA,
AUXOCYTES AND SPERMA-
TOCYTES.

GUSTAV EISEN, PH.D.

INTRODUCTORY.

THIS paper is a preliminary report of my investigations on the spermatogenesis of *Batrachoseps attenuatus*. The memoir will be published in the *Journal of Morphology*, Vol. XVII, No. 1. As, however, some considerable time must elapse before the paper can be published, it has been deemed proper to publish this short extract covering a few of the more important points.

Batrachoseps is adult in the months of June and July, and is at this time difficult to find, as it is then estivating deep below the surface. The testes were fixed by my iridium-chloride method and sectioned in paraffin. The stain used was the iron haematoxylin, according to Benda, and after-staining with congo. The sections were studied with Zeiss Apochromate, 2 mm. Ap. 1. 40. The light used was the incandescent gas-light passing through the achromatic filter described lately in the *Zeitschrift f. wiss. Microscopie*, Bd. XIV, p. 444.

The figures illustrating this paper are entirely diagrammatic. No special effort has been made to insert the correct number of chromioles in the chromosomes; and the author's want of skill in preparing this kind of drawing will account for many other discrepancies.

CONSTITUENTS OF THE CELL.

The following general division of the structures of the cells of the testes is proposed: Cytosome, Caryosome, and Archo-

some. This division is in accordance with the one proposed in my paper on the plasmocytes in the blood of *Batrachoseps*.¹

The cytosome comprises all that part of the cell situated exterior to the nucleus except the archosome. The cytosome contains the following distinct structures: cytoplasm proper, plasmosphere, hyalosphere, granosphere, metaplastic secretions, cytoplasmic membrane, and cell wall. None of these

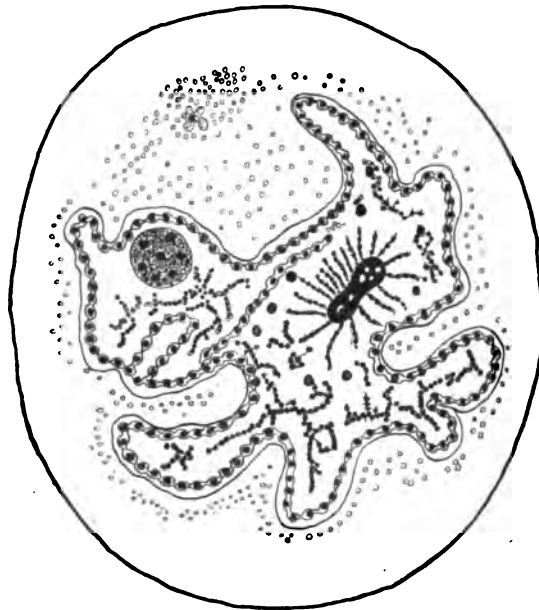


FIG. 1. — A polymorphous spermatogonium in the "perfect resting stage." The form of the nucleus allows the most perfect metabolism. Numerous chromioles are connected by a thread of chromoplasma. A network of linosomes is partially indicated, the individual granules being connected by Linopodia. A large chromoplast with endochromatic granules. Eight para-chromatic granules. A single archosome in the cytoplasm, the latter only partially indicated by small open circles. A single large linoplast, with seven endonucleolar granules.

structures are in any way intimately connected with the archosome. The three spheres mentioned above surround each other, like three concentric shells, at the time when the cell is in partial resting stage; but at a later stage, or as soon as the prophase is entered, these spheres break up and scatter in the cytoplasm proper. Each one of the spheres constitutes an independent structure, and they are not developed one from the other.

¹ *Proc. Cal. Acad. Sci.*, 3d Ser., Zoöl. Vol. i, No. 1.

The plasmosphere is the outermost sphere; the granosphere is the innermost one. The plasmosphere is the first one to break up into minor parts. These arrange themselves in the equatorial of the cell and serve as material for the mantle fibers

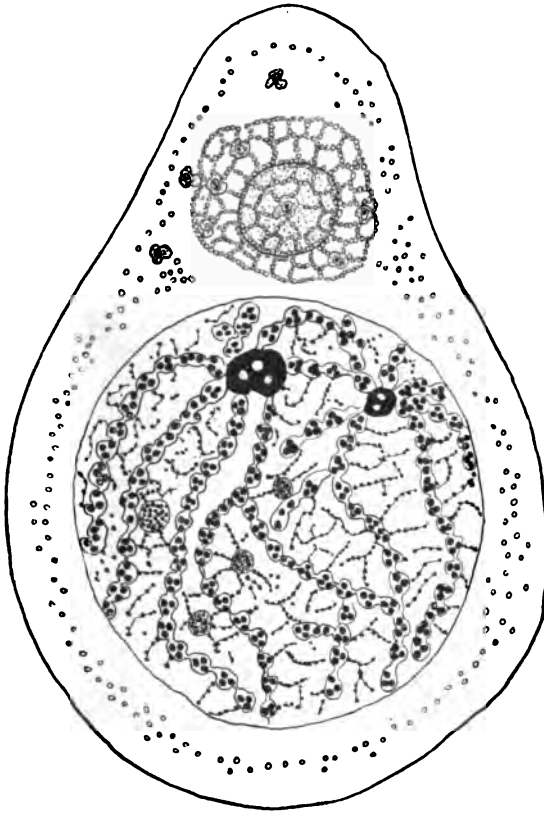


FIG. 2. — Auxocyte in the "imperfect resting stage," showing the formation of leaders consisting of round chromioles surrounded by a film of chromoplasm. The leaders start from two chromoplasts of unequal size, both containing endochromatic granules. The leaders are connected by a linosomic network. Four linoplasts. In the cytoplasm are seen the two spheres, the inner one, the granosphere, containing the archosome. There are eight accessory archosomes, some in the plasmosphere, others in the cytoplasm. The two spheres are of a foam-like structure. The cytoplasm is only partially indicated.

and for the new cell wall which is formed when the two daughter-cells separate.

The granosphere remains longer, but when it breaks up it furnishes material for the fibers of the central spindle. It also constitutes the main dwelling place of the archosome.

The archosome, or centrosomal structures, consists of three distinct parts, situated one interior to the other. These three parts constitute one single organized and individualized body — the archosome. The most interior part is the centriole ; this centriole is surrounded by a thin layer or zone — the somosphere. The somosphere is surrounded by a generally non-staining zone — the centrosphere. There are one or more centrioles.

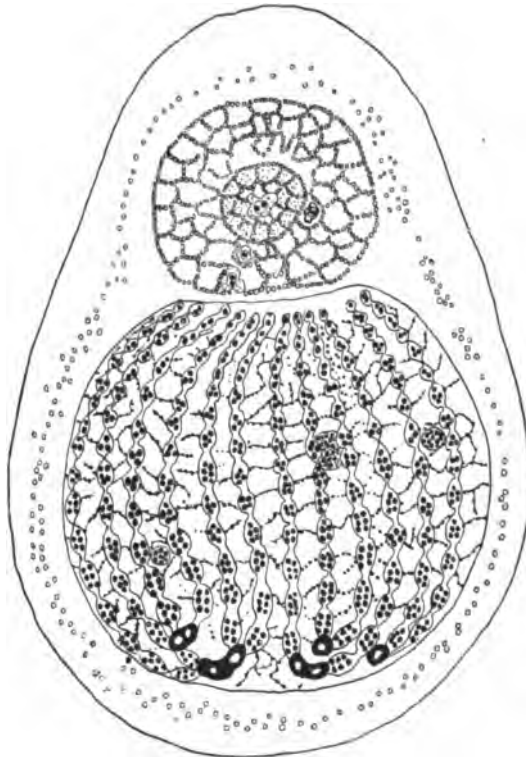


FIG. 3.—An auxocyte in the "bouquet stage." There are twelve leaders starting from five chromoplasts. The leaders consist of chromomeres containing chromioles suspended in a film of chromoplasm. The spheres are of a foam-like structure. There are three accessory archosomes and one archosome with two centrioles. The open space between the inner granosphere and the outer plasmosphere represents the hyalosphere. The cytoplasm is only partially indicated.

Both the somosphere and the centrosphere are amoeboid, especially the centrosphere. The latter constitutes the organ of locomotion of the archosome. There are besides the archosome several accessory archosomes. The function of the

archosome is to conduct the development and evolution of the fibers of the mitotic figures. The function of the accessory archosomes is to conduct the formation of the contractile fibers and perhaps furnish material for their construction. They also conduct the "fiber cones."

The location of the archosome is variable; it is sometimes situated in the granosphere, but is at other times found outside

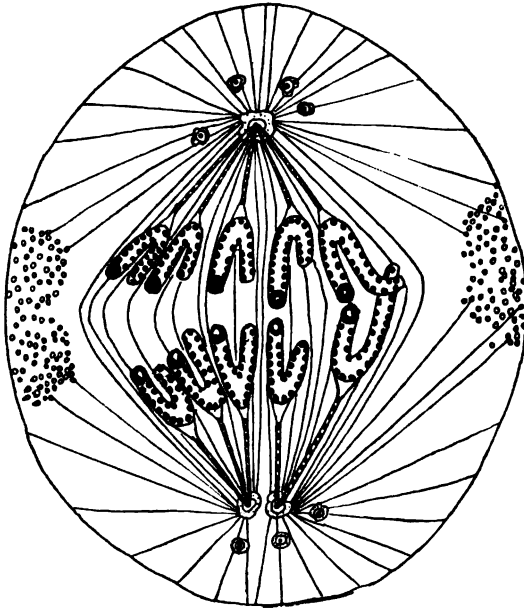


FIG. 4.—An auxocyte in the beginning of the anaphase. Only a few of the chromosomes are indicated. At each pole there are one and two archosomes and three and four accessory archosomes. The chromosomes contain chromioles suspended in chromoplasm. At the apex of each chromosome there is seen a chromoplast with endochromatic granules. To the right and left in the cell are seen agglomerations of plasmosphere indicating the position of the new cell wall, which is to separate the two daughter-cells. The chromosomes are seen to be connected with the chromioid by contractile fibers, the latter consisting of granules enclosed in a common sheath. The spindle fibers as well as the polar fibers start from the centrosphere.

of it. The accessory archosomes are of the same structure as the archosome, and one of the former may assume the function of the latter.

The accessory archosomes, if too numerous, are expelled from the cell, and then become paracellular bodies. Similarly, parts of the spheres are also expelled from the cell.

The nucleus contains the following more or less distinct

parts: chromioles, chromomeres, chromosomes, chromoplasts, linoplasts, linin, chromoplasm, endochromatic granules, and parachromatic granules.

Chromioles.—These are the most minute of the visible organized and individualized primary structures of the nucleus, and are the most important constituents of the chromosomes, probably being the carriers of heredity. They appear as minute globules, staining darker than the other parts of the nucleus except the chromoplasts. The chromioles are of a certain size and number in every species of nucleus and in every perfect chromosome. They are, as a rule, arranged in a regular manner in the chromosomes and in the chromomeres. During the absolute resting stage of the cell the chromioles are situated free in the nucleus, connected only by tiny filaments of linin and chromoplasm; while during the mitotic stages they are grouped into chromomeres, and these again into chromosomes. With absolute resting stage is indicated only absolute rest from "mitotic work." During this stage active metabolism is carried on.

There are thirty-six chromioles in every perfect chromosome, and these are divided among six chromomeres, each chromomere containing six chromioles. The chromioles are surrounded by a connective, apparently homogenous substance—the chromoplasm. The chromoplasm thus constitutes the greatest bulk of the chromosome.

The chromomeres are small aggregations of chromioles from three to six in number, according to the stage of development of the nucleus. The chromosomes in the polymorphous nuclei are twenty-four in number, but in the other testes cells there are only twelve. Each perfect chromosome contains six chromomeres.

In the resting stage of the polymorphous spermatogonia we find in the nucleus one or more large dark-staining bodies—the chromoplasts (net-knots). These chromoplasts are particular and most important organs of the nucleus. Their function is to attract the chromioles and to arrange them first into leaders, and later, through certain changes of the leaders, into chromosomes. The chromoplasts finally divide up into as many parts as there are chromosomes, one part adhering to each chromo-

some through its entire existence. The chromoplasts are characterized by one or more highly refractive endochromatic granules, which probably serve as nourishment for the chromi-

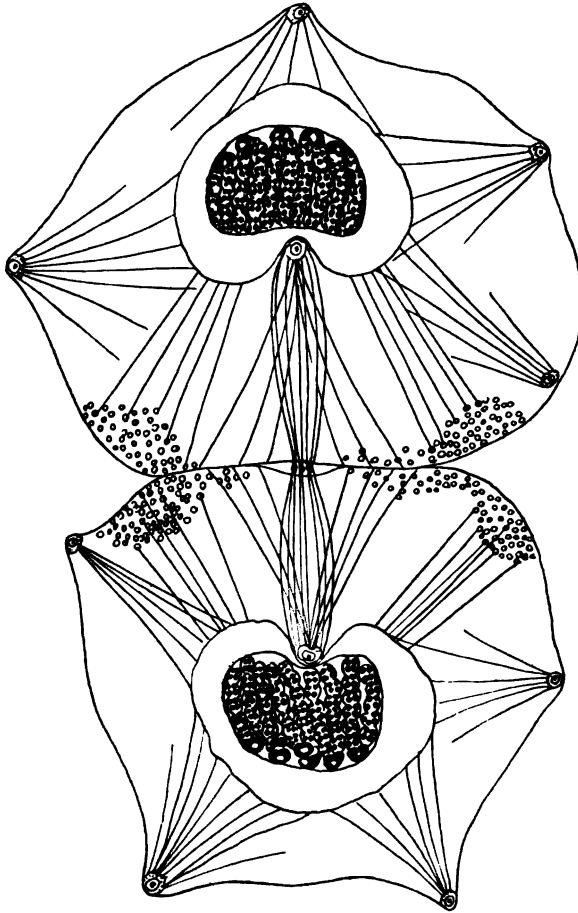


FIG. 5. — Two daughter-cells of an auxocyte connected by a spindle bridge. There are eight accessory archosomes at the apex of as many fiber cones. Two archosomes are connected by a central spindle. In the latter is seen a mid-body consisting of three condensation granules. The chromosomes are being regenerated, and the chromoplasts appear at the angle of the chromosomes instead of at the apex, as in the last cell stage. In one nucleus are seen five, in the other six chromoplasts with endochromatic granules. Between the true nuclear membrane and the false membrane is an open space caused by the false membrane being pulled away by the fiber cones.

oles. The chromoplasts serve as landmarks by which the position of the chromosomes can be ascertained with great accuracy.

The linin consists of minute granules — linsomes — arranged in a more or less regular network, which latter at certain times supports the elements of the chromosomes. The true nucleoli or linoplasts are principally agglomerations of linsomes, and serve as storage reservoirs for the linin network.

The nuclear membrane is formed apparently from linin and not from cytoplasm proper. During the anaphase a false nuclear membrane is formed from cytoplasm proper, but this membrane is again dissolved as soon as the object for which it is formed is accomplished.

SPINDLES AND SPINDLE FIBERS.

The following varieties may be segregated: mantle fibers, polar fibers, central spindle fibers, contractile fibers, retractile fibers, and fiber cones. All these fibers originate only in connection with an archosome. The contractile fibers alone are directly connected with the centriole of the archosome. All

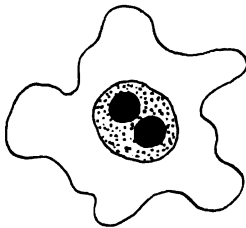


FIG. 6. — An archosome consisting of an outer centrosphere, an inner somosphere, with two centrioles.

the other fibers and rays emanate from the outer margin of the centrosphere of the archosome, but do not penetrate into this sphere, and accordingly are not connected with the centrioles or the somosphere. The material for the mantle fibers is furnished from the granules and the secretions of the plasmosphere; while the material for the central spindle is furnished by the granules and secretions of the granosphere.

The contractile fibers are those which connect the chromosomes with the archosome. They are from the beginning of a different structure from any of the other fibers, being beaded and highly contractile. Their structure strongly recalls that of muscle fiber.

The fiber cones are particular structures, so far not met with in any other cells. They consist of bundles of fibers held together at one point by an accessory archosome, while the distal ends of the fibers are attached to the false nuclear membrane

formed around the nucleus at the time of the anaphase. The archosome moves away and carries with it the fibers, which pull away the false nuclear membrane, thus causing a vacuole to form around the nucleus. The object of all this is probably to enable the nucleus to develop without the interference of surrounding structures. These fiber cones are frequently very numerous, as many as seventeen having been counted in a single cell. They are of large size and cause the cell membrane to be pushed out.¹

The spindle bridge, which connects two or more cells, consists of the remnant of the central spindle. As the spindle bridge exists only in cells which commence the same phase of mitosis at the same time, it is probable that the purpose of the spindle bridge is to time or regulate the commencement of this mitosis. The mid-body of the spindle bridge serves probably as a storage reservoir for the cytoplasm of the spindle bridge.

VARIETIES OF CELLS.

The testes of *Batrachoseps* contain four distinct varieties of cells, as follows : polymorphous spermatogonia, auxocytes, spermatocytes, and spermatids. These originate one from the other in the order mentioned above. Of these varieties there are one or more generations. They are characterized as follows :

Polymorphous Spermatogonia. — These possess a perfect resting stage in which the nucleus is polymorphous as regards form, being greatly indentated and folded during the perfect resting stage. The nucleus during this stage contains neither chromosomes nor chromomeres, the chromioles being scattered about and not connected with the chromoplasts. These cells give rise to several generations of cells of the same nature, with the exception that there is no perfect resting stage like the one in the mother-cell, and that consequently the nucleus is not folded, but perfectly even, round, or oblong. The mitosis of the poly-

¹ Fiber cones of similar appearance, but of a different nature, have been described by botanists from the pollen cells of higher plants. These cones, however, do not possess archosomes.

morphous spermatogonia and their offspring is through twenty-four chromosomes and somatic division. The last generation of these cells gives rise to the auxocytes.

The auxocytes are characterized as follows: They possess a bouquet stage; they are the first cells with twelve chromosomes; their mitosis is heterotypic and by equation; they pos-

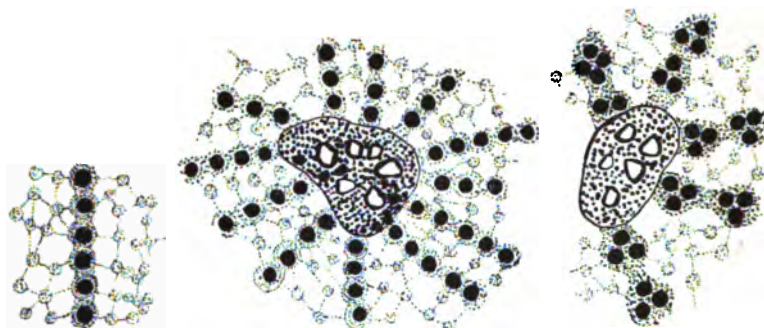


FIG. 7.

FIG. 8.

FIG. 9.

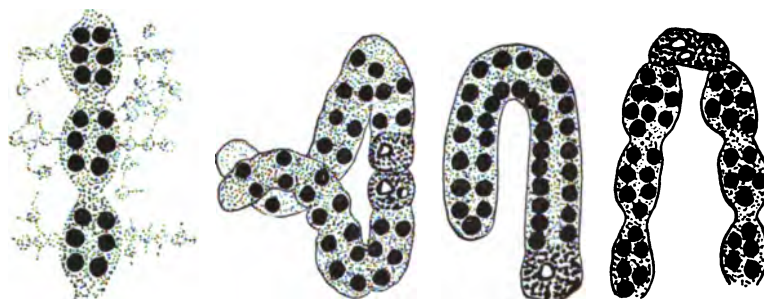


FIG. 10.

FIG. 11.

FIG. 12.

FIG. 13.

FIGS. 7-13 represent a broken series of leaders illustrating the formation of the leader and the chromosome.

FIG. 7.— Isolated row of chromioles surrounded by chromoplasm and suspended in a network of linozomes.

FIG. 8.— Chromoplast with twelve leaders of chromioles. From the imperfect resting stage of the polymorphous spermatogonium.

FIG. 9.— Chromoplast with five leaders. Each leader is made up of chromomeres, and each chromomere consists of three or more chromioles surrounded by chromoplasm. A network of linozomes between the chromomeres.

FIG. 10.— Three chromomeres, each with six chromioles surrounded by a chromoplasm and suspended in a network of linozomes.

FIG. 11.— A pretzel chromosome containing chromioles and two chromoplasts with endochromatic granules.

FIG. 12.— A chromosome from the metaphase. It contains thirty-six chromioles and a terminal chromoplast with an endochromatic granule.

FIG. 13.— Part of a chromosome from the spermatocyte.

sess no perfect resting stage, the chromioles being arranged into leaders and always connected with the chromoplasts; there is but one generation; the daughter-cells are the spermatocytes; and they have numerous fiber cones at the end of the anaphase.

The spermatocytes are characterized as follows: They have numerous fiber cones in the beginning of the mitosis, homotypic mitosis with twelve chromosomes and with equation division; no bouquet stage and no perfect resting stage; but one generation; the daughter-cells are the spermatids which give rise to the spermatozoa through direct development and growth; and the chromosomes are *V*-shaped.

THE MITOSIS.

The mitosis is the result of two distinct and separate processes which, for the greater part, run parallel and independent of each other, but which meet at certain nodes in order to



FIG. 14.—A diagrammatic representation of the structure of the granosphere. The dotted globules are cytoplasmic granules, and between them are seen metaplastic secretions represented by small open rings. The globules are connected by Linopodia, and form a foam structure, partly a network.

accomplish certain objects jointly. These processes are the chromosomic process and the radiosomic process.

The radiosomic process is presided over by the archosome and the accessory archosomes, and consists in the development and evolution of the various fibers and the central spindle, in the evolution of the spheres, and the dissolution of the nuclear membrane. To this process belong also the development and dissolution of the false nuclear membrane and the reabsorption of the fibers.

The chromosomic process is presided over by the chromo-

plasts, and consists in the development and evolution of the leaders out of chromoplasm and the chromioles, the formation of the latter into chromomeres and chromosomes, and the multiplication of the chromioles and their proper distribution in the chromosomes. The two processes coöperate in the separation and equation of the chromosomes, which coöperation commences with the dissolution of the nuclear membrane. To the chromosomic process belongs also the movement of the chromoplasts in the umbrella-shaped and confluent nucleus at the end of the anaphase. With this process the archosomes have

nothing to do, as it is accomplished before the nuclear membrane is dissolved by the mantle fibers.

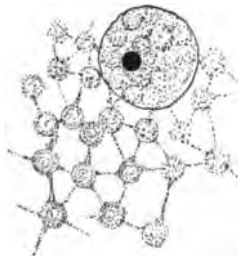


FIG. 15. — A diagrammatic representation of the structure of linosomes and the linoplast. The individual linin granules are connected by means of Linopodia. The linoplast contains linosomes as well as an endonucleolar body.

The radiosomic process commences with the dispersion of the spheres. The plasmosphere is dispersed first, and its granules are arranged in the equatorial of the cell, there to furnish material for the new cell walls. The central spindle fibers are then formed out of material furnished by the granosphere, which is in this way entirely used up. The nuclear membrane is dissolved by the mantle fibers and not by the

central spindle fibers. The contractile fibers are formed after the central spindle fibers have reached considerable size.

The chromosomic process begins with the formation of leaders out of chromioles and chromoplasm. The chromioles aggregate into chromomeres, and, later on, a certain number of these form chromosomes. Their formation is shortly as follows: The leaders to the number of twelve are connected with the chromoplasts, and by contraction and a certain arrangement assume the bouquet stage. The leaders then split lengthwise, the two forks being held together by a fragment of the chromoplasts. The chromoplast divides into as many parts as there are to be chromosomes, but each part is always attached to a leader. Next, the two halves of the leader spread apart and twist around each other and thus form a pretzel-shaped chromosome. By this time the nuclear membrane is dissolved and

the pretzel-shaped chromosomes are placed on the central spindle, where they are taken hold of by the contractile fibers, which attach themselves to the prongs halfway between the

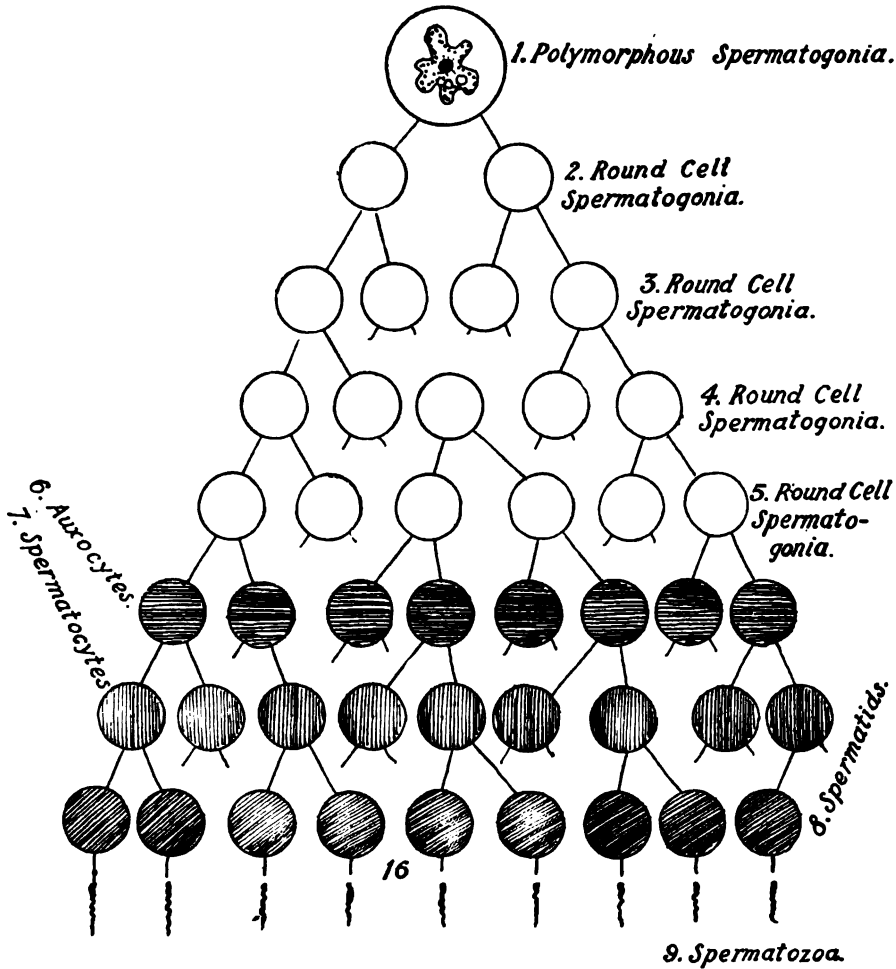


FIG. 16. — Diagram of the cell generations in *Batrachoseps* testes; 1, polymorphous spermatogonia; 2 to 5, four generations of round cell spermatogonia; 6, auxocytes; 7, spermatocytes; 8, spermatids; 9, spermatozoa.

chromoplasts and the free end. Each half is then pulled away and the chromosome is formed by an equation and not by a reduction. In the new chromosome the fragment of chromo-

plast is attached to one of the ends. This process is the one that takes place in the auxocytes.

The next step is the formation of a confluent umbrella stage or ring-like nucleus. The object of this form is to allow the chromoplasts to change their place. When the nucleus is reorganized in the spermatocyte the chromoplasts are found to be situated not at the end of each chromosome, but at the angle of the fork. This change of position could not take place except through the medium of an umbrella-shaped nucleus. During this stage the chromioles are also doubled. The nucleus now passes through a stage of growth which is facilitated through the large vacuole which is formed around the nucleus with the aid of the fiber cones and the accessory archosomes.

In the spermatocyte the central spindle is frequently formed from two opposite fiber cones left over from the last mitosis. The chromosomes of the spermatocytes are *V*-shaped before mitosis. They are divided longitudinally in the way usual in the homotypic mitosis, and by equation, not by reduction. During the prophases of the radiosomic mitosis the superfluous archosomes are expelled from the cell and remain for some time as paracellular bodies between the cells.

PERMANENCY AND NATURE OF THE CELL STRUCTURES.

The cytosome proper contains no permanent structures of any kind. The plasmosphere, hyalosphere, granosphere, the various kinds of fibers, as well as the central spindle, are all ephemeral structures which are developed by rearrangement of preëxisting granula, and which again disperse when their function is over. The granula contained in the cytosome is at least of four different kinds, and everything points to the conclusion that one kind of granula is never converted into any other kind. In other words, the granula of the granosphere is not evolved from the granula of the plasmosphere, etc., but both are independent and individualized primary structures as compared with the secondary ones of spheres and fibers. For the principal granula of the cell the following terminology is proposed: cytosomes, plasmosomes, hyalosomes, somosomes, granosomes, and lino-somes, the latter being of nuclear origin.

If we turn to the nucleus, we find similarly that the chromomeres, the chromosomes, and the leaders are also ephemeral and secondary structures which form and disperse, the chromioles alone being the permanent individualities of the chromosomic structures. The nucleus then contains the following permanent granula: linosomes, the chromoplasmic granula, the chromioles, and the granula composing the chromoplasts. The permanent structures of the cell are the centrioles, the chromioles, and the chromoplasts. As regards the primary parts of these last-mentioned structures we are yet in doubt, but there is every reason to believe that these structures are of a highly complicated nature.

Similarly the fibrillar and alveolar structures of the protoplasm are only secondary, ephemeral, and temporary. With proper optical means we see that the alveole, as well as the reticulum, is built up of granules. These granules adhere to each other by means of minute projections or arms, for which I have proposed the name of Linopodia. The ultimate visible structure of the protoplasm is thus a granule, capable of projecting and retracting Linopodia.

For a fuller explanation and demonstration of these facts I must refer to the larger paper now in the hands of the publisher of the *Journal of Morphology*.

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ERRATUM.

In No. 1, p. 1, 13th line, last word, read *ganglion* in place of "*gland*."

BIOLOGICAL BULLETIN.

THE EARLY CLEAVAGE AND FORMATION OF THE MESODERM OF SERPULORBIS SQUAMIGERUS CARPENTER.

S. J. HOLMES.

THE material upon which the present paper is based was collected at San Pedro, Cal., in the summer of 1895. Work upon it was carried on for a time during the winter of 1895-96, under Prof. C. O. Whitman, at the University of Chicago; but as the series of stages the material afforded proved incomplete, the subject was laid aside in the hope that, at some future time, when new material could be collected, the gaps might be filled. Since it is improbable that an opportunity of remedying this defect will soon present itself, and as the development of this form shows several interesting points of comparison as regards the formation of the mesoderm with what has recently been found to obtain in other mollusks and certain annelids, it was thought best to publish the present account. The development of *Vermetus*, a genus from which *Serpulorbis* is somewhat doubtfully distinct, has been studied by Salensky¹ in considerable detail. According to Salensky, mesoblastic pole cells do not appear, and the mesoderm in *Vermetus* arises at a comparatively late period of development by a proliferation from the ectoderm in the region of the blastopore. With this conclusion my own observations do not agree, as certain stages that were found afford very clear evidence that the mesoderm arises

¹ *Archives de Biologie*. I, vi, 1887.

from the posterior macromere *D*, as has been found in so many other cases among mollusks and annelids.

Serpulorbis squamigerus is a common mollusk on the coast of southern California. The shell early loses all traces of its originally spiral form, and becomes bent and twisted in a very irregular way. Many individuals are often found tangled together, resembling a group of worm tubes, and forming masses of considerable size. The eggs are deposited in elongated capsules attached by one end a short distance within the mouth of the shell. In addition to the eggs the capsules contain numerous small cells, probably follicular, which doubtless serve to nourish the developing embryos. A large number of the eggs in each capsule fail to develop normally, and sooner or later break up into masses of isolated blastomeres. The cleavage of such

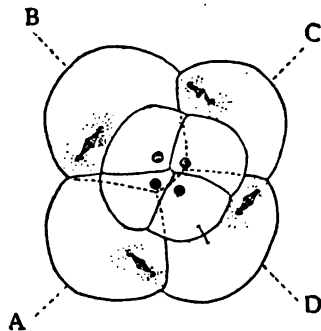


FIG. 1. — Eight-cell stage, seen from the animal pole. The dextrotropic origin of the first quartette of micromeres is indicated, and the spindles in the angles of the macromeres show that the next division will be laetotropic.

eggs is irregular, sometimes from the start, but often the irregularity appears only after the egg has developed for some time in an apparently normal manner. As this departure from the typical path of development occurs at different stages in different eggs, it is not always easy to distinguish the normal from the abnormal process of cleavage.

The first two cleavages are total and equal, giving rise to a four-cell stage of the usual molluscan type, in which two cells meet in a cross furrow at the vegetal pole. The next cleavage, which results in the formation of the first generation, or quartette, of micromeres, is dextrotropic. The micromeres are rather small, as is the rule in molluscan eggs, in which, as in the present case, there is a large amount of yolk. At the next cleavage the second quartette of micromeres are given off from the macromeres in a laetotropic direction. The spindles appear at one angle of the macromeres, but before the next division the nuclei wander through the cell so that the spindles next

appear at the opposite angle. The same migration is repeated in an opposite direction in preparation for the next ensuing division. The appearance of the second quartette is soon followed by a laetotropic division of the cells of the first. A dextrotropic cleavage of the second quartette next appears, and at about the same time the macromeres bud off the third quartette in a right-handed spiral, completing the separation of the ectoderm from the entoderm. The twenty micromeres composing the ectoderm are all transparent and devoid of yolk. While they form about one-half the surface of the egg, they

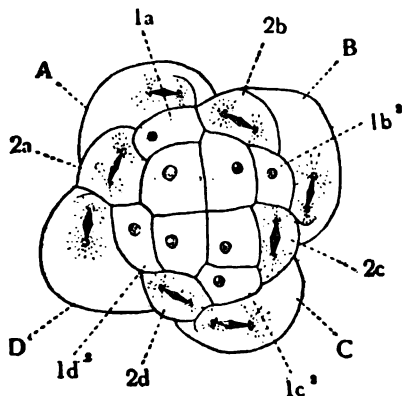


FIG. 2.

FIG. 2. — Sixteen to twenty-four cell stage from the animal pole, showing the origin of the third quartette and the dextrotropic cleavage of the second. The first quartette has divided, producing the four "trochoblasts."

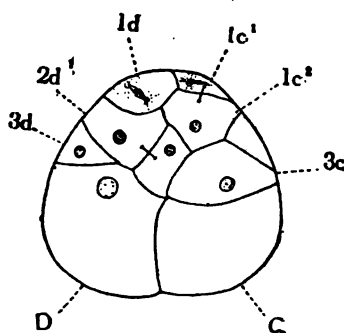


FIG. 3.

FIG. 3. — Lateral view of the twenty-four cell stage. A cleavage is taking place in the apical cells.

form much less than half its bulk, as their thickness is not nearly so great as that of the large yolk-laden entomeres. The conclusion drawn by Salensky, that in *Vermetus* there are four quartettes of micromeres produced, is doubtless erroneous. The cleavage of the first quartette of ectomeres was probably overlooked, and the outer products of this division regarded as having had a separate origin from the macromeres. A comparison of Salensky's figures with the cleavage of *Serpulorbis* renders this interpretation probable. Besides, there are strong reasons for doubting that four generations of ectomeres are ever produced among the gasteropods, as I have attempted to show elsewhere.

The next cleavage occurs in the macromere *D*, and results in the formation of a yolk-laden cell, lying obliquely above the larger stem cell in such a way as to indicate that the division was laeotropic. This cell corresponds exactly as regards its time and mode of origin with the primary mesoblast cell of other mollusks. The corre-

sponding division of the other three macromeres to form the remainder of the fourth quartette does not occur until a considerably later period. These divisions do not give rise to ectomeres, but to large yolk-laden entomeres, the cells of the fourth quartette being somewhat larger, if anything, than those at the vegetal pole.

About the time the primary mesoblast cell is given off the four apical cells of the first quartette divide in a dextrotropic direction, the outer products of the division forming the basal cells of the arms of the cross. Up to this time the cleavages of *Serpulorbis* agree, point for point, with those of *Crepidula*, *Lymnaea*, *Limax*, *Planorbis*, and *Physa*, with the exception that the divisions in the latter two genera are reversed. A comparison of

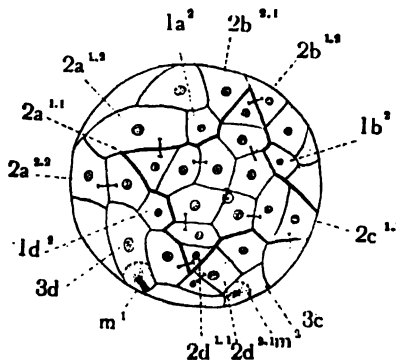


FIG. 4. — Forty-eight-cell stage, seen from the animal pole. The outline of the cross is marked with a heavier line. A dextrotropic twist is apparent in the arms of the cross. The small mesoblast cells are shown in dotted lines on the posterior side of the egg.

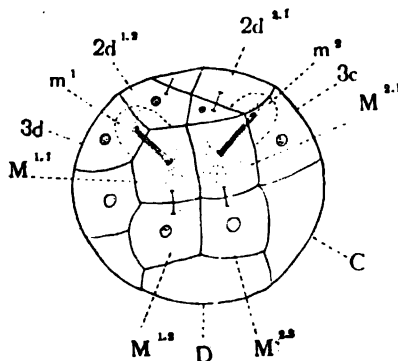


FIG. 5. — Posterior side of the same egg, showing the four derivatives of *4d*, the upper pair budding off the mesomeres, *m*¹ and *m*², into the interior of the egg.

the forty-eight-cell stage, shown in Figs. 4 and 5, indicates that the following divisions have taken place: The four upper cells of the second quartette have divided in a laeotropic direction, giving rise to the cells *2a*¹⁻¹, *2b*¹⁻¹, etc., which

form the tip cells of the arms of the cross. The four tip cells are smaller than the others, as in *Crepidula* and *Planorbis*. A cleavage of the four lower cells of the second quartette has taken place, likewise in a laeotropic direction. The second quartette now contains sixteen cells in four groups of four cells each. These cells bear exactly the same relations to each other, to the arms of the cross, and to the adjacent cells of the other quartettes, that they do in *Crepidula*, *Planorbis*, and several other forms at the corresponding stage of development. There can be no doubt, therefore, of their derivation, though their actual divisions were not all observed. The large entomeres, *A*, *B*, and *C*, have divided laeotropically, completing the formation of the fourth quartette. In place of the mesoblast cell 4 *D* there is a group of four cells, an upper pair containing little yolk, and a lower pair of about the same size in which the yolk is abundant. The origin of these cells was not followed, but there can be little doubt that they all owe their origin to the mesoblastic pole cell. They occupy the same area that was occupied by the pole cell. The cap of ectodermic cells is radially symmetrical, and contains no cells of sufficient size to have given rise to such large cells as the upper pair of the four without altering very materially the symmetrical relations shown in the figure. Besides, nothing corresponding to such a division is seen in other forms. In all probability these cells arose first by a horizontal division of the mesoblast cell, such as occurs in a large number of forms, and then by a division of the two daughter-cells in a plane at right angles to the previous one. At this period the egg contains a regular cap of ectodermic cells, four entomeres at the vegetal pole, three entomeres, 4 *a*, 4 *b*, and 4 *c* of the fourth quartette, and the group of four cells above described, in place of the remaining cell of the fourth quartette, 4 *d*. A comparison with the corresponding stage in the egg of *Crepidula*, as shown in Fig. 31 of Conklin's paper,¹ shows that the cells of the ectodermic cap correspond point for point, and also that the four cells we have derived from 4 *d* are represented in *Crepidula* by four cells of subequal size having the same origin and position. The fourth quartette is formed a

¹ *Journ. Morph.* Vol. xliii. 1897.

little later in *Crepidula* than the stage shown in Fig. 31, otherwise the two eggs are practically identical. The four cells in *Serpulorbis* are all on the surface of the egg and are not overlapped so much by the ectomeres

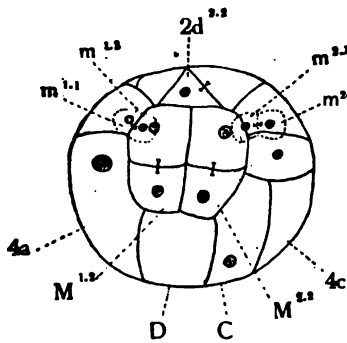


FIG. 6.—View of the posterior side of an egg in a somewhat later stage. There are seen two pairs of mesomeres in the cleavage cavity.

as in *Crepidula*. The upper pair in Fig. 4 is shown in process of division. Each cell buds off a small cell into the interior of the egg, the spindles diverging anteriorly. At about the same period an almost exactly similar division occurs in *Crepidula*, the upper pair of cells budding off a small cell into the interior of the egg in very nearly the same direction. At a somewhat later stage in *Serpulorbis*, shown in Fig. 6, I have found two pairs of small cells lying entirely within the cleavage cavity which probably represent the descendants of the single pair whose origin has just been described. The parallelism with *Crepidula* extends also to this division as the corresponding pair of small cells in that form divides at about the same period. This is as far as the cleavage of these cells was carried. These results were worked out before Conklin's paper appeared, and as I did not follow the further history of these cells, as Conklin has succeeded in doing in *Crepidula*, it seemed uncertain what interpretation of them should be made. It seemed improbable that all of 4d should form the mesoblast, as it was supposed to do in several forms. The four large cells showed no signs of passing into the interior of the egg, and it is probable that, after the mesoderm is separated from the upper pair, they enter into the formation of the entoderm, as in *Crepidula*.

lapped so much by the ectomeres as in *Crepidula*. The upper pair in Fig. 4 is shown in process of division. Each cell buds off a small cell into the interior of the egg, the spindles diverging anteriorly. At about the same period an almost exactly similar division occurs in *Crepidula*, the upper pair of cells budding off a small cell into the interior of the egg in very nearly the same direction. At a somewhat later stage in *Serpulorbis*, shown in Fig. 6, I have found two pairs of small cells lying entirely within the cleavage cavity which probably represent the descendants of the single pair whose origin has just been described. The parallelism with *Crepidula* extends also to this division as the corresponding pair of small cells in that form divides at about the same period. This is as far as the cleavage of these cells was carried. These results were worked out before Conklin's paper appeared, and as I did not follow the further history of these cells, as Conklin has succeeded in doing in *Crepidula*, it seemed uncertain what interpretation of them should be made. It seemed improbable that all of 4d should form the mesoblast, as it was supposed to do in several forms. The four large cells showed no signs of passing into the interior of the egg, and it is probable that, after the mesoderm is separated from the upper pair, they enter into the formation of the entoderm, as in *Crepidula*.

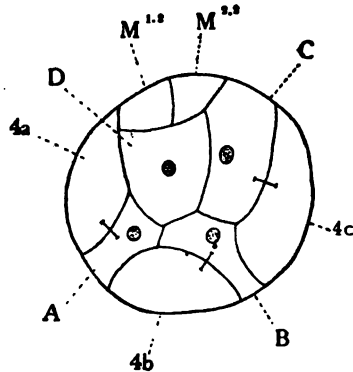


FIG. 7.—Vegetal pole of the same egg shown in Fig. 5.

Recent researches render it probable that the cell 4*d* is not typically a purely mesoblastic cell. As Conklin has pointed out, the divisions of 4*d* in *Umbrella* are strikingly like those in *Crepidula*, and strongly indicate, as Conklin maintains, that this cell contains both mesoderm and entoderm. The same cell in *Cyclas* was held by Stauffacher¹ to produce both mesoderm and entoderm. In *Unio*, Lillie² found that the pole cells budded off small cells at the surface before giving rise to the mesoblastic bands, and among annelids similar phenomena have been observed by Mead³ in *Amphitrite*, by Wilson⁴ in *Nereis* and *Aricia*, and by Treadwell⁵ in *Podarke*. In *Planorbis*⁶ I have found that a minute cell is budded off from each of the pole cells before they divide to form the mesoblastic bands. It has been suggested by Wilson that these minute cells correspond to the small entomeres found in *Nereis*, but they are budded off in a different direction and lie in the cleavage cavity instead of on the surface. This does not prove, however, that they are not homologous with entodermic cells, as a slight change in the direction of division of the pole cells would bring them in the wall of the blastula. And as they are probably not functional they may represent the last vestige of the entodermic portion of the mesentomeres.

¹ *Jen. Zeit.* Bd. xxviii. 1893.

² *Journ. Morph.* Vol. x. 1895.

³ *Journ. Morph.* Vol. xiii. 1897.

⁴ *Ann. N. Y. Acad. Sci.* Vol. xi. 1898.

⁵ *Biological Lectures*, delivered at Woods Holl, Session of 1898, 1899.

⁶ *Zoöl. Bull.* Vol. i. 1897.

NEW SPECIES OF HYGROCELEUTHUS AND
DOLICHOPUS, WITH REMARKS ON
HYGROCELEUTHUS.¹

AXEL LEONARD MELANDER AND CHARLES THOMAS BRUES.

THE recognition of two new species of *Hygroceleuthus* and a study of both sexes of the other American species of this genus, and of another species which has been hitherto placed in *Dolichopus*, have shown the necessity of revising this genus. Hitherto but little attention has been paid to the females, which are very difficult to separate, whereas the males present very evident characters and are easily identified.

Previous to 1868 only one species of *Hygroceleuthus* was known from North America, and three others from the rest of the world. Since then North America has produced at least eight species, making it the richest country known in species of this genus.

Hygroceleuthus and *Dolichopus* are very closely allied, their separation being effected by male characters alone. These two genera form a group distinct from other *Dolichopodidae* by the presence of a number of bristles on the upper surface of the hind metatarsi. They have in common also the first joint of the antennae hairy above, third joint short, its arista dorsal, and hypopygium free.

The so-called distinction between the two genera is to be found in the length of the face which, in the typical males of *Hygroceleuthus*, is lengthened and attains the lower corner of the eye. Subordinate to this and even less constant are the lengthened antennae, deep incision in the hind margin of the wing, and broadened wings. In the three typical species of *Hygroceleuthus*, which have tarsal ornamentation, this occurs on the middle legs. In *Dolichopus* there is no species with the

¹ Contributions from the Zoological Laboratory of the University of Texas, under the direction of W. M. Wheeler, No. 1.

middle legs similarly ornamented if we except *plumipes*. For this reason and because it shows a tendency toward the lengthened face of *Hygroceleuthus*, we have included *plumipes* in the present paper. But as this species shows strong *Dolichopus* characters in the short, stout antennae and slight costal thickening, it cannot be placed satisfactorily in either genus as they

have been defined. On the other hand, the European *Hygroceleuthus diadema* merges with *Dolichopus* on account of its shortened antennae.

The original definition of *Hygroceleuthus* included a deep incision in the hind margin of the wing and broadened wings. From these characters *Aldrichii* and *Wheelerii* deviate very decidedly.

Latipes, the only North American *Hy-*

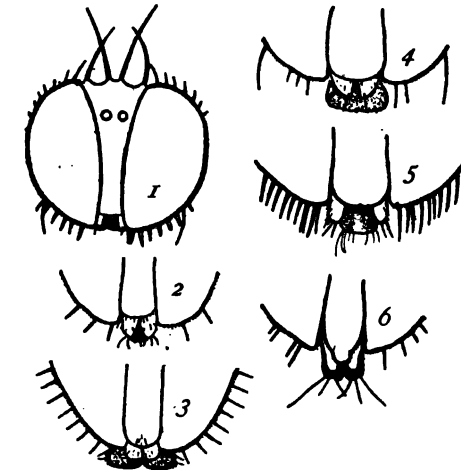


FIG. 1.—Showing length of face: 1, *Dolichopus comatus*, male; 2, *Hygroceleuthus plumipes*, male; 3, *Hygroceleuthus Wheelerii*, male; 4, *Hygroceleuthus amnicola*, female; 5, *Hygroceleuthus afflicta*, male; 6, *Hygroceleuthus latipes*, male.

groceleuthus which Loew saw, possessed no characters at variance with the typical species. It was because of limited material that Loew felt justified in constructing this genus. Like other genera founded on secondary sexual characters alone, such as *Rhagoneura* and *Spathochira* of this same group, *Hygroceleuthus* has been found invalid as the number of species increased.

From the foregoing it seems advisable that *Hygroceleuthus* be no longer retained with generic value, but may be kept as an expression for a group of the genus *Dolichopus*.

Of the previously described species of *Hygroceleuthus*, one has failed to be recognized, *lamellicornis* Thom., if indeed this be a species of *Hygroceleuthus*. The type was a female from California, but the description omitted the important points.

We have examined types of all the species except *latipes*, *plumipes*, *crenatus*, *afflictus*, and *ciliatus*. The specimens studied in the preparation of this paper are in the collection of Dr. Wm. M. Wheeler, who kindly placed his collection at our disposal.

Although the name *ciliatus* has been previously used by Walker,¹ Aldrich's *ciliatus* may remain, as Walker's species is too poorly characterized to admit of its recognition.

Males.

Middle tarsi ornamented	2
Middle tarsi plain	5
2. Antennae largely black	<i>Aldrichii</i> Wheeler
First joint of antennae yellow	3
3. Middle tarsi strongly compressed	<i>latipes</i> Loew
Middle tarsi not compressed, first joint feathered laterally	4
4. Middle tibia twice length of femur	<i>Wheelerii</i> , sp. nov.
Middle tibia not elongated, slender	<i>plumipes</i> Scop.
5. Cilia of tegulae yellow	6
Cilia of tegulae mostly black	8
6. Second abdominal segment laterally with a tuft of yellow hairs	<i>afflictus</i> O. S.
Abdomen without such tuft	7
7. Face yellowish white	<i>crenatus</i> O. S.
Face silvery	<i>idahoensis</i> Aldrich
8. Arista bare	<i>ciliatus</i> Aldrich
Arista densely pubescent	9
9. Front coxae yellow, postocular cilia in part yellow	<i>consanguineus</i> Wheeler
Coxae black, postocular cilia wholly black	var. <i>propinquus</i>

Females.

First joint of antennae yellow	2
First joint of antennae in great part black	4
2. Species about 6 mm. first joint of middle tarsus yellow at base	3
Species about 4 mm. Middle tarsi wholly black	<i>plumipes</i> Scop.
3. Hind tibiae wholly yellow, vertex green	<i>latipes</i> Loew
Hind tibiae black at tip, vertex violet	<i>latipes</i> var. <i>cognatus</i>
4. Tip of hind tibiae black, or, if yellow, the wings narrow	5
Hind tibiae wholly yellow	6

¹ List of Diptera in Collection of British Museum, pt. iii, p. 661.

5. Front femora with the basal two-thirds infuscated *amnicola* sp. nov.
Front femora wholly yellow *Aldrichii* Wheeler
6. Arista with slight pubescence; wings usually with a stump-vein at the bend of the fourth vein 7
Arista bare 8
7. Second joint of hind tarsi yellow at base; legs yellow; smaller species *crenatus* O. S.
Second joint of hind tarsi black; legs darker; larger species *consanguineus* Wheeler
8. Tegular cilia wholly black, somewhat robust . . . *ciliatus* Aldrich
Tegular cilia yellow at sides 9
9. Wings yellowish anteriorly, coxae yellow . . . *afflictus* O. S.
Wings hyaline, coxae darker *idahoensis* Aldrich

Hygroceleuthus Wheelerii, sp. nov.

Male. Length 5 mm.; length of wing 4 mm. Shining metallic cupreous green. Proboscis piceous. Face covered with a thick dust, silvery on lower half, becoming golden towards antennae. Antennae yellow, first two joints wholly so, the third black on upper surface and outer half. First joint hairy above, and with a slight swelling on inner surface to meet the other antenna; second joint tipped with a fringe of black hairs, becoming

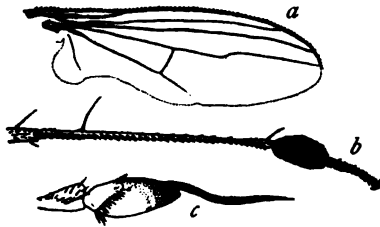


FIG. 2.—*H. Wheelerii*: a, wing of male; b, middle leg; c, antenna.

stouter and longer on underside, nearly one-half the length of first joint when viewed from above. Third joint somewhat longer than the first, bearing dorsally a stout arista with very short pubescence. Vertex metallic violet. Postocular cilia delicate, black above and light yellow below. Thorax bright grassy green, becoming cupreous at sides and with a faint indication of the two narrow approximated median brown lines.

Abdomen green, with silvery dust at sides and beneath. Posterior margins of segments becoming cupreous and margined with piceous. Hypopygium green, almost piceous, overlaid with a grayish dust. Lamellae pale, with a distinct narrow dark border and a black fringe. Internal appendages yellow. Sides of thorax glaucous; shining green when viewed from behind. Fore coxae yellow, hairy on whole anterior face and with a few bristles at tip. Middle and hind coxae yellow with outer face glaucous at basal two-thirds. Trochanters, femora, and tibiae yellow. Middle tibiae very long and thin, the proportion of femur to tibia of the middle leg being 20 to 39. Hind tibiae not incrassate, nor with smooth space on inner surface. Anterior tarsi black from tip of first joint, middle and hind tarsi black. Middle

tarsi short, first joint broadly feathered laterally. Wings narrow, hyaline, distinctly yellowish towards costa. The usual costal swelling at tip of first vein is slight. Almost no incision at tip of fifth vein. The anal angle of wing is produced into a large distinct lobe. Veins dark. Bend in fourth vein regular. Halteres and tegulae yellow, the tegular cilia long and black.

One male specimen taken by Dr. Wm. M. Wheeler in a cranberry bog at Woods Holl, Mass., July 13, 1899.

This very distinct species is readily recognized by its lengthened middle tibiae. Aside from this the following are more or less characteristic: the reduced costal swelling and incision of the wing as well as the pronounced anal lobe; the peculiar lateral ornamentation of the middle tarsi, which are unusually short; the violet front; the light-colored antennae and finely pubescent arista; and the yellow hind tibiae.

Hygroceleuthus plumipes Scopoli.

Male. Length 3.5-4.5 mm. Length of wing 3.5-4 mm. Face yellow pollinose. Antennae yellow, third joint black at tip. First joint with a slightly prominent projection on its inner side. Arista slightly pubescent. Front metallic green. Thorax without distinct dusted bands. Abdomen

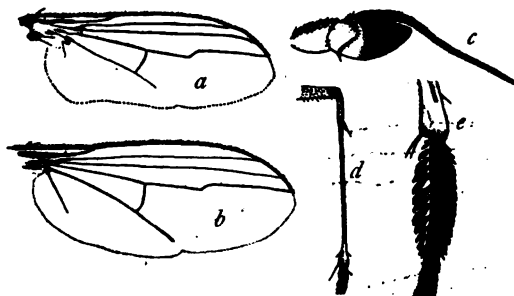


FIG. 3. — *H. plumipes*: a, male wing; b, female wing; c, antenna; d, middle tibia; e, middle metatarsus, male.

metallic green above and distinctly bronzed toward the apex; white dusted at the sides and covered throughout with short black hairs. Lamellae of hypopygium narrowly bordered with fuscous. Pleurae metallic green, covered with white dust. Coxae of same color as the pleurae, except the anterior ones, which are yellow and covered with black hairs on the anterior and inner surfaces, bearing also a few black bristles at their tips. Femora yellow. Tibiae, yellow, the middle pair slightly, and the posterior pair distinctly tipped with black. Middle tibiae flat, very slender except at extreme

base and apex, which are normal in form. The flat sides each with a wide, shallow, piceous groove extending along the entire length of the tibia. Tarsi black, except basal two-thirds of anterior pair. Middle tarsi with the first joint longer than the two following and broadly feathered laterally. Wings narrow, the anterior and posterior margins subparallel, nearly hyaline. Swelling at tip of humeral vein slight, incision at tip of fifth vein slight. Tegulae with long black cilia.

Female. Length 3.5–4.5 mm. Length of wing 3.5–4 mm. Face broader, gray, greenish in certain lights and darker below. Middle tibiae and tarsi of the usual form. Anal lobe of wing more rounded than in the male, and the costa not thickened.

Twenty-three specimens examined. Sixteen males and six females, from Rabbit Ear Pass 10,000 feet, and North Park, 9000 feet, Colorado. Also one male specimen from Vancouver Island, collected by Mr. C. Livingston.

This species is readily distinguished by the peculiarly formed middle tibia and tarsus of the male. The female may be separated from *latipes* by its smaller size and wholly black middle tarsi, and from all the other species by the entirely yellow first antennal joint.

The distribution of this species is most interesting. It is one of the three species of *Dolichopus* which are common to Europe and North America. It is mostly a boreal species, being found in great numbers throughout Northern Europe, from Cape North to Switzerland. In America it was noticed by Loew from Alaska. Where *plumipes* extends toward the south it is limited to high altitudes, as witnessed in Switzerland and Colorado.

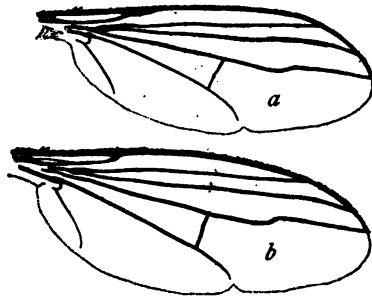


FIG. 4. — *H. latipes*: a, male; b, female.

Hygroceleuthus latipes Loew.

Male. Length 5–7 mm., of wing 4.5–6.5 mm. Face silvery, yellowish above. First joint of antennae yellow, at most slightly darkened above, long. Arista pubescent. Vertex generally green. About 6 to 8 of the supraocular cilia black, the remainder pale. Abdomen with posterior margins of the segments cupreous. Lamellae of hypopygium white with narrow black border and fringe. Anterior coxae yellow, hairy on distal

portion in front. Femora and tibiae yellow. Middle tarsi compressed, ornamentation dorsal on last four joints. Wings thickened at tip of first vein and incised at fifth. Tegula cilia black, a few yellow inside.

Female. Face silvery, broader. Antennae shorter, first joint hairy above, sometimes infuscated above. Vertex green. Abdomen more cupreous, and anterior and middle tarsi slightly lighter than in the male. Posterior femora with two macrochaetae near tip on outer side. Wing incision not very deep.

This species has a greater distribution than any of the other species, except *plumipes*. It has been taken at various places in the Northern States from Connecticut to Idaho. This is the commonest species, and, aside from *Wheelerii*, the only species yet found east of the Dakotas.

Latipes, var. ? *cognatus*. Two specimens vary from the type as follows and may possibly represent another species. Posterior tibiae black at tip and hind tarsi totally black. Vertex violet. Posterior femora each with only one macrochaeta on outer side near apex. One female from Woods Holl, Mass., July 19, 1899, and another female from Pullman, Ill., August 7, 1897.

Hygroceleuthus Aldrichii Wheeler.

Male. Length 4-5 mm. Face with silvery white dust below, ochreous above. Antennae black, first and second joints yellow below on mesial surface. Arista moderately pubescent. Front green. Postocular cilia white on lower two-thirds, black above. Lamellae of hypopygium yellow with black border and fringe of delicate black hairs. Anterior coxae yellow, others dark. Second, third, and fourth joints of middle tarsi distinctly compressed and fringed with stout black hairs. Anal angle of wing bilobed, costal thickening prominent and incision at tip of fifth vein slight. Tegulae with long black cilia.

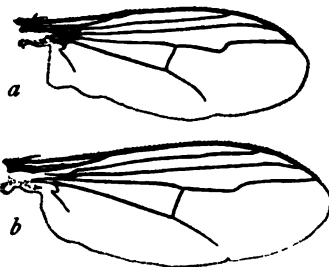


FIG. 5.—*H. Aldrichii*: a, male; b, female.

Female. Length 4-5.5 mm. Face grayish-yellow. First joint of antennae almost entirely black. Tip of hind tibiae usually black. Incision at tip of fifth vein slight. Anal angle not bilobed, and tarsi but very slightly compressed.

Numerous specimens examined, males and females. From Idaho, Wyoming, and Colorado.

The peculiar anal lobe of the male wing easily identifies this species. The female is not so easily distinguished, but can be recognized by the characters given above.

Hygroceleuthus amnicola, sp. nov.

Female. Length 4.5 mm., of wing 4.5 mm. Of a bright metallic green with cupreous reflections. Palpi light yellow with black hairs. Face evenly overlaid with golden dust. Antennae black with lower half of first and second joints yellow. The difference in color is sharply marked. First joint hairy above, with a rather large yellow projection from inner side. Second joint tipped with a fringe of black hairs which are longer below. Front metallic brassy green. Upper half of the postocular cilia black, lower pale. Thorax shining green, not much dusted in front, disc somewhat cupreous;

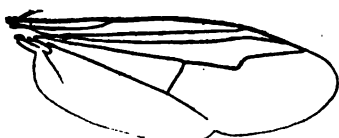


FIG. 6.—*H. amnicola*: wing of female.

the two narrow approximated lines are left green. Sides of thorax glaucous, becoming more piceous in all the coxae. Front coxae with black hairs on whole anterior face. Middle and hind femora yellow; fore femora black for nearly proximal

two-thirds. All the tibiae yellow, infuscated at tip; the darkening especially prominent on the hind legs. Front tarsi black from tip of first joint; middle tarsi with first and second joints yellow, their tips black, remaining joints black; hind tarsi black from base of first joint. Wings long and narrow, greatly prolonged beyond tip of fourth vein; the fourth vein with a very strong bend and continued obliquely forward. Halteres and tegulae yellow, the cilia of the latter long and black.

One specimen, Colorado, Grizzly Creek, North Park; collected by Mr. C. F. Baker.

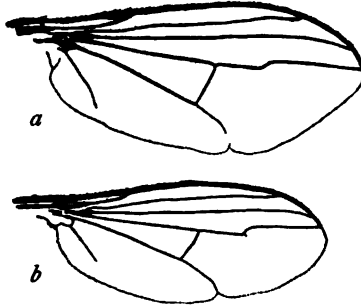
Although this species is represented by a single female specimen, it is so distinct that there is no hesitancy about its position. The wings reach further beyond the fourth vein; the angle of the fourth vein is more nearly rectangular; the coxae are darker and the femora blacker than in any other female *Hygroceleuthus*.

Amnicola differs from *Aldrichii* thus: middle tarsi are not compressed and are largely yellow; the front femora and coxae are much darker; the wings are hyaline and more extended beyond the veins, and the fourth vein is more sharply bent.

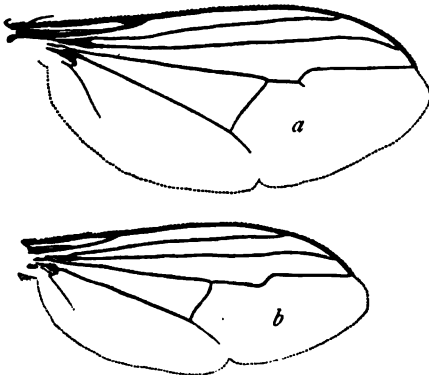
Hygroceleuthus crenatus O. S.

Male. Length 5-6 mm. Face yellowish-white. Antennae with the two basal joints black, except a yellow protuberance on the inner side of each. Arista densely pubescent. Postocular cilia black on upper third, yellow below. Anterior coxae yellow, with a black stripe outwardly. Femora and tibiae yellow; the hind tibiae incrassate, with a shallow, broad, brownish groove on the inner side. Anterior and middle tibiae infuscated toward the tips. Hind tarsi black except base of first joint. Lamellae of hypopygium nearly white, margined with black at the tips. Wings very broad, narrowed to the base. Costa moderately thickened, incision at tip of fifth vein moderate. Cilia of tegulae yellow, delicate, sometimes with a few black hairs intermixed.

Female. Length 5-6 mm. Face uniform gray. First joint of antennae in great part black. Arista black, slightly pubescent. Hind tibiae wholly yellow, the hind tarsi with the second joint yellow at the base. Wings with a distinct incision at tip of fifth vein. A stump-vein projecting from the bend of the fourth vein, sometimes abbreviated.

FIG. 7.—*H. crenatus*: a, male; b, female.

Numerous male and female specimens examined from California, Washington, Wyoming, Idaho, and Vancouver Island.

FIG. 8.—*H. consanguineus*: a, male; b, female.*Hygroceleuthus consanguineus* Wheeler.

Male. Length 5.5-6.5 mm., of wing 4.5-5.5 mm. Upper two-thirds of face more opaque than lower third, generally with two broad vertical bands on upper two-thirds. Antennae black, in small part yellow below, and on mesial surface of first and second joints. First joint with smooth swelling inside. Arista thick, densely pubescent. Postocular cilia black, becoming thick and flat below; upper infraorbital cilia bright orange, lower black. Lamellae of hypopygium

piceous with suffused black border. Legs yellow, black from tip of first tarsal joint. Hind tibiae incrassate slightly. Distal portion of fourth vein with abrupt angle and with stump-vein. Cilia of tegulae black.

Female. Somewhat smaller and with relatively longer wings. Stump-vein at angle of fourth longitudinal present. Tegular cilia black. Fore coxae with black hairs in front. The dilation of first antennal joint is less prominent. The lower postocular cilia are also parti-colored but less flattened than in the male.

This species was described from a large number of specimens collected in July, 1896, near Monterey, Cal.

Consanguineus, var. *propinquus*. Several interesting specimens received from Mr. C. Livingston, from Corfield, Vancouver Island, vary from the typical *consanguineus* as follows:

Darker. All the coxae piceous; femora piceous beneath near base. Postocular cilia black, none of the orange-colored cilia of the typical *consanguineus* present, not so many of the infraocular cilia flattened. Lamellae of hypopygium darker.

Hygroceleuthus afflictus O. S.

Male. Length 6-6.5 mm. Face white, silvery. Antennae with yellow expansion on inner side of first joint; second joint with only a vestige of yellow on the inner side. Pubescence of arista sparse but robust. Vertex green. Postocular cilia black above for a long distance, descending nearly to the middle of the eye; below light yellow. Second abdominal segment bearing on each side near the middle a tuft of long yellow hairs, directed backward and reaching to the middle of the fourth segment. Third segment with a very small similar tuft. Hind tibiae incrassate, with a broad shallow groove on the inner side. Costal thickening and incision at fifth vein of wing distinct.

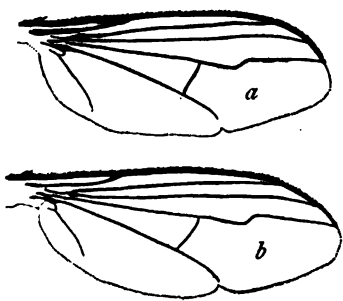


FIG. 9. — *H. afflictus*: a, male; b, female.

Female. Length 5.5-6.5 mm. Face gray, with a greenish tinge on the lower part and slightly ochreous near the base of the antennae. Antennae dark, first and second joints in great part black.

Arista bare. Abdomen without any tufts of yellow hair. Anterior coxae yellow, sometimes with a small posterior stripe dark. Hind tibiae completely yellow. Wings yellowish anteriorly, costa not thickened, notch at tip of fifth vein very pronounced. Tegular cilia black, yellow at the sides.

Numerous males and females examined from Arizona, Monterey County, Cal., and Wyoming. It was described from San Rafael, Cal., and is recorded also from Washington.

The male of this species is very easily known by the presence of the tufts of yellow hair upon the second abdominal segment.

Hygroceleuthus ciliatus Aldrich.

Male. Length 4-5.5 mm. Face yellowish-white. Front green. Antennae black, except lower half of first and second joints. Arista bare. Post-ocular cilia black on upper third, below nearly white. Sides of first abdominal segment with a few white hairs. Tips of hind tibiae blackish. Tarsi simple, black from tip of first joint. Wings narrow, hyaline, costa not thickened at tip of first longitudinal. Indentation at tip of fifth vein slight. Tegulae with long black cilia.

Female. Length 4-5.5 mm. Face yellowish-gray. Arista of antennae bare. Hind tibiae wholly yellow. First joint of hind tarsi lighter at base. Tegular cilia black. Wings with a distinct incision at tip of fifth vein.

Numerous specimens examined from South Dakota and Wyoming.

Hygroceleuthus idahoensis Aldrich.

Male. Length 5.2 mm., of wing 4.8 mm. Face silvery. Antennae black, not large but with swollen yellow protuberance on inner side; second joint slightly yellow on inner side; arista rather stout. Vertex blue-green. Lamellae of hypopygium small, white, with rather wide black margin. Anterior coxae yellow with a dark green stripe on outer face, and with a few hairs on lower part. Hind tibiae incrassate with a longitudinal depression. Tarsi black from tip of first joint. Costa thickened for a long distance, the incision in hind margin slight. Tegular cilia pale, not large.

Female. Face broader, darker than in the male. Anterior coxae more hairy. Wings less yellow anteriorly, costa not thickened. Tegular cilia larger, black with a slight admixture of pale ones.

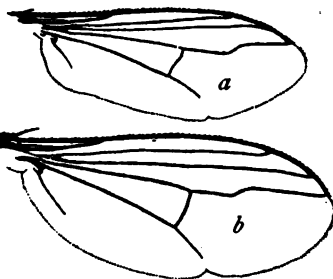


FIG. 10.—*H. ciliatus*: a, male; b, female.

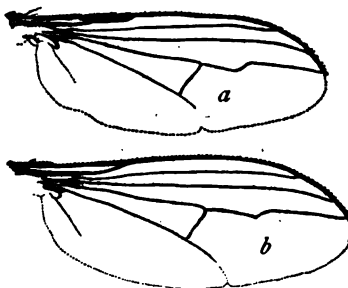


FIG. 11.—*H. idahoensis*: a, male; b, female.

Moscow, Idaho. September. The original collection numbered about seventy-five specimens.

LIST OF THE SPECIES OF THE GROUP HYGROCELEUTHUS.

- plumipes* Scopoli, 1763. *Ent. Carn.*, 334.
latipes Loew, 1861. *Neue Beitræge*, Fasc. viii., 5.
 ? *lamellicornis* Thomson, 1868. *Eugenies Resa*, 511.
crenatus Osten Sacken, 1877. *Western Diptera*, 312.
afflictus Osten Sacken, 1877. *Western Diptera*, 313.
ciliatus Aldrich, 1893. *Kan. Univ. Quart.*, 25.
idahoensis Aldrich, 1894. *Kan. Univ. Quart.*, 154.
Aldrichii Wheeler, 1899. *Proc. Cal. Acad. Sci.*, 3.
consanguineus Wheeler, 1899. *Proc. Cal. Acad. Sci.*, 5.
Wheelerii Melander and Brues, *sp. nov.*
amnicola Melander and Brues, *sp. nov.*

Dolichopus.

The following notes and descriptions were made from specimens belonging to Dr. Wm. M. Wheeler, who has not only given us his entire collection to work over, but has also tendered us much aid and advice.

The appended list is given in the hope that it may prove useful, as it contains many new localities. It is interesting to note that so many of Loew's species have been again recognized.

Dimorphism has not been noticed in the genus *Dolichopus* as yet, but a most interesting case of what may turn out to be such is to be found in the species *Henshawii* and *marginatus*. Of the more specific characters these two species possess in common the following: antennae similarly colored, vertex violet, fore coxae with dark hairs, hind tibiae with similar dark glabrous stripes, similar wing neuration, and the yellow hind femora of the male ciliated with black hairs, in which character they differ from all other dolichopodes. On the other hand, the males seem evidently distinct as follows:

Henshawii. Face generally yellow; postocular cilia darker yellow; fore tibiae incrassated at tip; fore tarsi ornamented and banded; hind tibiae not evidently darkened towards tip

except a large black blotch on inner side; lamellae of hypopygium fringed with comparatively short hairs.

Marginatus. Face gray; all the legs plain; front tarsi gradually darker toward tip; hind tibiae more infuscated at apex; lamellae fringed with numerous longer hairs.

The females of these species cannot be separated. They agree rather with *marginatus* in the color of the postocular cilia and of the legs. The males, evidently so distinct, were taken, together with the females, in the same netful at Woods Holl, Mass., by Dr. Wheeler. *Marginatus* is the commoner form. In all were taken from July 14 to August 9, 1899, forty-eight females, thirteen male *Henshawi*, and nineteen male *marginatus*.

Dolichopus partitus, sp. nov.

Femora chiefly black, cilia of inferior orbit black, wings infuscated, coxae wholly black.

Male. Length 5-5.5 mm., of wing the same. Dark green with metallic lustre. Proboscis and palpi black. Face rather wide, short, concave beneath the antenna, and with a pronounced transverse ridge at its lower fourth, below this convex. Face covered with light brown pollen, except a small spot at each side of the ridge. Antennae totally black; the first joint with short bristles above; the bristles about the apex of the second joint much longer below. Third joint short, ovate, obtusely pointed at tip; arista black, pubescent. Front dark violaceous green. Postocular cilia totally black. Thorax above, dark green, with a median longitudinal dark cupreous band. Scutellum of same color as thorax. Abdomen metallic green, lighter than thorax. Surface covered with short black

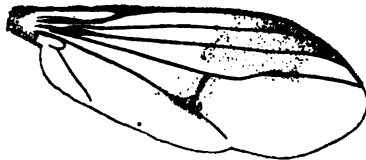


FIG. 12. — *D. partitus*: male wing.

hairs, more sparse towards base; very slightly covered with whitish dust. Hypopygium almost black, shining with two patches of black hair on dorsal side near the base; internal appendages ferruginous. Lamellae yellow, of usual-size, with a black border. Between the white center and black border is a ferruginous band. The border is very much jagged at apex and furnished with strong bristles, becoming more slender towards base. Pleurae greenish-black, covered with whitish dust; coxae black. Legs black, except femora and tibiae just at their articulation, the four anterior tibiae and the base of the first joint of four anterior tarsi. Posterior femora not ciliated. Wings infuscated about cross-vein and at apex between costa and third vein. The

latter spot reaches only to the second longitudinal in one specimen. Veins black; costa with an elongate swelling at the junction of the humeral vein; notch at tip of fifth vein distinct. Tegulae and halteres light yellow, the former with long black cilia.

Described from two male specimens collected in North Park, Colorado.

This species is related to *Johnsoni* Aldrich, but may be distinguished by its wide face, totally black coxae, spotted wings, and violaceous front.

Dolichopus paluster, sp. nov.

Bluish-green; antennae totally black; infraocular cilia black; tegular cilia black; legs including coxae black; tarsi not ornamented; hind femora ciliated in male.

Male. Length 5-5.5 mm. Wing 4.5-5 mm. Shining bluish-green. Proboscis and palpi piceous. Face moderately wide, between three and four times as long as the width at the middle, covered with brownish-yellow pollen, not at all silvery. Vertex dark blue-green. Postocular cilia all black. Antennae totally black; first joint with but few bristles above, those about the apex of the second joint very long below. Third joint oval, obtuse at apex. Arista black, pubescent, about twice as long as the antenna. Dorsum of thorax dark green, tinged with blue. In some specimens there is a median stripe, more blue and shining. Scutellum of the same color as thorax, fringed with short light-brown hairs. Abdomen green, distinctly bluish in many specimens, and very shining, sharply compressed towards apex

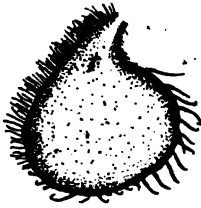


FIG. 12.—*D. paluster*: male lamella.

and somewhat inflated near the base; destitute of light dust. Hypopygium black, shining, slightly ochreous-dusted near the base, and bearing a bunch of black hairs basally. Lamellae oval, slightly angulated inwardly, nearly white, with a sharply defined black border, fringed with black bristles which are more delicate basally. Internal appendages dark ferruginous. Pleurae black, white dusted, those of the prothorax green like the dorsum. Legs, including coxae, wholly black, fore coxae white dusted, and with short black hairs. Anterior tarsi not ornamented, about one-fourth longer than the tibiae; middle tarsi but slightly longer than tibiae. Hind femora ciliated on apical half with black hairs, the longest hairs not longer than the width of the femur at the point of their insertion. Posterior tibiae somewhat thickened. Wings grayish; veins black; costa but slightly thickened at tip of first longitudinal; fourth vein not sharply bent, approximated with the third vein at tip. Incision at tip of fifth vein slight. Tegulae and halteres yellow, tegular cilia black.

Female. Size same. More coppery than the male, especially on the sides of the thorax and abdomen. Face dark yellowish-gray; slightly more than twice as long as wide. Posterior femora not ciliated below, hind tibiae not thickened. The wings are brownish, darker anteriorly between the costa and second longitudinal; the veins black, very narrowly margined with brown. Otherwise like the male.

Described from five male and four female specimens, collected by Dr. Wm. M. Wheeler, in Monterey County, Cal., during July, 1896.

This species is most closely related to *corax* Osten Sacken, from which it differs as follows: lamellae nearly white, bordered with black; fore tarsi male plain. In *corax* the front tarsi are ornamented and the lamellae are nearly black, yellowish-brown in the middle only.

Dolichopus intentus, sp. nov.

Femora largely black; tibiae pale; cilia of inferior orbit dark; tegular cilia dark; wings hyaline; lamellae of hypopygium small, dusky; antennae black, third joint long, pointed, with subapical arista.

Male. Length 4 mm., of wing 3.5 mm. Dark bronzed green dusted. Proboscis dirty yellow, palpi piceous. Face thickly covered with silvery dust, except a small median spot immediately below antennae. Antennae black; first and second joints subequal; first two joints more or less shining, densely clothed with appressed short pubescence; third joint more opaque, the pubescence closer. First joint bristly; second joint with a terminal fringe of bristles which become longer beneath; third joint longer than first and second together. Arista subterminal, shorter than third antennal joint. Front violet, metallic, slightly bronze dusted. Postocular cilia black. Thorax and abdomen greenish-bronze above, becoming piceous dusted below. Hypopygium piceous dusted, shining inwardly. Internal appendages dark; lamellae small, fuscous without a distinct darker border, fringed with hairs only. Legs plain, dark, with usual bristles. Front coxae somewhat lighter than pleurae, yet silvery. Femora piceous except the yellow tip; hind femora with two ante-apical bristles. Fore and

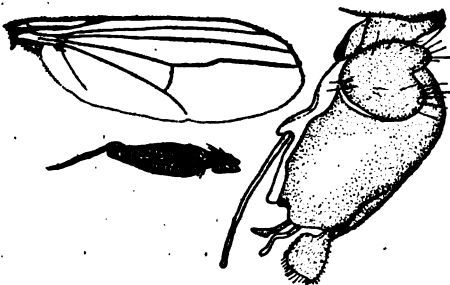


FIG. 14. — *D. intentus*: male wing, antenna and hypopygium.

middle tibiae yellow ; hind tibiae black at tip, slightly swollen along middle, but without a smooth space internally. Wings subquadrate, hyaline, third and fourth veins subparallel at tip. Wings with costa at tip of first vein thickened and without an obvious notch at terminus of fifth vein ; anal angle rounded. Tegulae and halteres yellow. Tegular cilia black.

One specimen, collected by Dr. Wm. M. Wheeler at Chicago, Ill., dated May 8, 1896.

This species is allied to *laticornis* Loew, and *incongruus* Wheeler, but is at once distinct in the structure of the antennae.

In his table of *Dolichopus*,¹ Mr. Aldrich commits *incongruus* to the section with the femora yellow. The type specimen has dark legs. Division 5 of his table may be thus altered :

- | | | |
|-----|--|----|
| 5. | Third joint of antennae large | 5a |
| | Third joint as usual, tegular cilia black | 6 |
| 5a. | Tegular cilia yellow ; hind tibiae dark on whole under surface | |
| | <i>incongruus</i> Wheeler | |
| | Tibiae of hind legs infuscated towards tip | 5b |
| 5b. | Tegular cilia generally yellow ; lamellae of hypopygium clear | |
| | <i>laticornis</i> Loew | |
| | Tegular cilia black ; lamellae of hypopygium dusky | |
| | <i>intentus</i> nov. | |

Dolichopus calainus, sp. nov.

Femora chiefly black, cilia of inferior orbit pale, middle tibiae black, femora yellow only at extreme tip, hind femora not ciliated, legs wholly black.

Male. Length 5 mm., of wing 4.5 mm. Bright metallic blue with greenish reflections. Proboscis and palpi piceous. Face of usual length and rather narrow ; light gray below, ochreous and darker above. Antennae totally black, third joint ovate, obtusely pointed at tip. Arista black, moderately pubescent, nearly twice as long as the antenna and inserted about the middle of the third joint. First joint but slightly bristly above, more strongly so toward the tip. Front bright blue with a decided greenish tinge. Postocular cilia black above, below the middle light. Just before the lower corner of the eye they are suddenly somewhat longer and placed very close together, forming a sort of brush. Dorsum of thorax and scutellum deep shining blue, greenish only at extreme sides and in front. Abdomen much compressed toward the apex ; shining bluish-green, whitish dusted on the sides below and covered with black hairs, which grow longer toward the apex of the abdomen. Hypopygium piceous, with several conspicuous

¹ *Kan. Univ. Quart.* Vol. ii., No. 1, p. 2.

patches of black hairs; internal appendages light brown. Lamellae small, strongly infuscated, lighter at middle; with a narrow black border which is much wider on the lower corner; fringed with black bristles which are

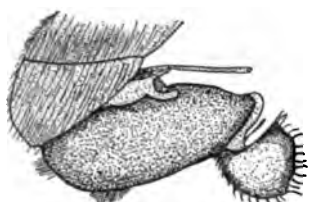


FIG. 15.—*D. calaisus*: hypopygium.

slender, especially on the upper edge. Pleurae very dark green, grayish dusted. All the coxae black. The anterior ones silvery in front and covered with short black hairs. Legs black, slightly whitish dusted. The anterior tibiae dark brown on the inner side. All the femora at extreme tip, the tibiae at extreme base and the first joint of anterior and middle tarsi at extreme base, yellow.

Wings hyaline, the veins black. Costa with a knot-like swelling at junction of humeral vein. Tegulae and halteres yellow, the former with long black cilia.

Described from one male specimen collected by Dr. Wm. M. Wheeler in Chicago, May 8, 1896.

This species is related to *myosota* O. S., but may be distinguished by the lamellae of the hypopygium, which are larger, darker, wider, and distinctly angulate below.

Dolichopus enigma, sp. nov.

Dark green, shining; wings brownish in front; tegular cilia black; cilia of inferior orbit pale; femora black, hind pair of male not ciliated; fore tibiae brownish-yellow; lamellae of hypopygium subrectangular.

Male. Length 4 mm., of wing 3.5 mm. Bright green, not very shining. Proboscis and palpi piceous. Face rather wide, covered with dense silvery dust, brownish in certain lights. Antennae totally black, sericeous, but little hairy above. First joint long, second and third taken together, about twice the length of first. Arista less than twice as long as antenna, black, but little pubescent. Front dark green, not very shining. Postocular cilia black above and pale below. Dorsum of thorax and scutellum bright green, somewhat cupreous in front. Abdomen dark green, bronzed, not so bright as thorax; covered with black hairs throughout and white dusted on sides and below. Incisures between segments black. Hypopygium

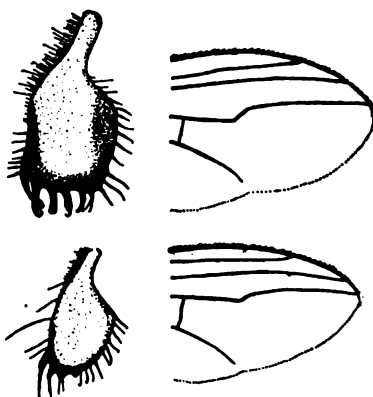


FIG. 16.—*D. enigma*, male; *D. ovatus*, male.

black, basal portion opaque, white dusted, with two patches of black hair dorsally; towards the apex very shining. Internal appendages ferruginous. Lamellae subrectangular, dirty, translucent, white, with brown border, wider at apex, where it is jagged and bristly. Pleurae very dark green, opaque, white dusted. Legs, including coxae, totally black, except the anterior tibiae above and the base of anterior tarsi, which are more or less yellow above, femora indistinctly tipped with brownish-yellow. Tarsi not ornamented, hind tarsi with the usual bristles. Wings grayish, tinged with brown in front and along the veins; costa with a short swelling in the angle which it makes with the first vein; bend in the fourth vein not very abrupt; second and third veins much approximated except at tip; no distinct incision at tip of fifth vein. Tegulae and halteres yellow; tegular cilia black.

One male, North Park, Colorado, over 9000 feet, collected during July.

This is closely related to *ovatus* Loew, but is distinct by the much larger subrectangular lamellae, costa with a swelling, second and third veins more approximated, and wings brownish in front.

Dolichopus agronomus, sp. nov.

Femora chiefly black, cilia of inferior orbit pale, middle tibiae yellow, first joint of hind tarsi with few bristles, hind femora ciliated with short hairs.

Male. Length 3.5 mm., of wing 3 mm. Dark metallic green. Proboscis and palpi piceous. Face very long, densely covered with bright silvery pollen, which continues past the antennae as far as the frontal bristles.

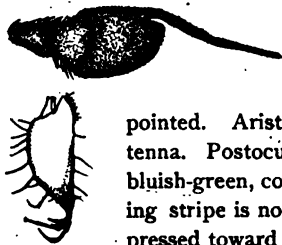


FIG. 17. — *D. agronomus*: male antenna and lamella.

Above the antennae it is greenish-white and not so dense. Antennae long, totally black, the first two joints short, the third large and broad, elongated ovate and rather sharply pointed. Arista black, pubescent, a little longer than the antenna. Postocular cilia black above, pure white below. Thorax bluish-green, covered with very fine white dust. A median shining stripe is not at all dusted. Abdomen very strongly compressed toward apex, dark green, white dusted, especially along the sides. The extreme basal and apical margins of the segments more or less free from the dust. Entire abdomen covered with short black hairs. Hypopygium black, shining, covered at base with white dust. Internal appendages light yellow. Lamellae nearly white with an indistinct narrow blackish border; elongate oval. Each lamella nearly bilaterally symmetrical, but little angulate inwardly and beset with the usual bristles. Pleurae greenish-black, dusted with gray. Coxae of same color as the pleurae, all tipped with yellow, the

anterior ones silvery in front. Femora brownish-black, tipped with yellow. Anterior and middle tibiae yellow, the anterior ones lighter. Posterior tibiae and tarsi deep black, the former yellow at extreme base. Anterior and middle tarsi blackened from the tip of first joint. Wings oval, much narrowed toward the base, hyaline, the veins dark brown. Costal swelling and incision at tip of fifth vein not well marked. Tegulae and halteres yellow. Tegular cilia yellow, with a couple of strong black ones intermixed.

Described from one male specimen, collected by Dr. Garry deN. Hough, at New Bedford, Mass., June 8.

From *convergens* it differs by the vertex being white pollinose, as well as the face. Also the hind femora are ciliated with short hairs; the hind tibiae are totally black; the lamellae of the hypopygium are oval, and the third and fourth veins of the wing converge less strongly.

From *albiciliatus* it differs by the smaller size; longer third antennal joint, and the black hind tibiae. Moreover, the ciliation of the hind femora of the male is shorter; the lamellae are not broad and rounded, and are much lighter in color.

From *xanthocnemus* it can be readily distinguished by the shorter ciliation of the hind femora and the black hind tibiae.

This is a very peculiar species and superficially resembles the species of the group *Hygroceleuthus*, although it is otherwise quite different.

Dolichopus pernix, sp. nov.

Green; face whitish; antennae black, arista plain; infraocular cilia white; tegular cilia black; feet yellow, including fore coxae, tip of hind tibiae conspicuously black; last two joints of male fore tarsi moderately enlarged, black; fourth longitudinal vein not broken.

Male. Length 4.75 mm., of wing 4.5 mm. Green, shining. Proboscis piceous, palpi yellow. Face narrow, silvery white, flavescent towards antennae. Antennae wide, black, first joint dark brown below; joints subequal; second and third together ovate; third obtusely pointed; arista dorsal, sericeous, longer than antenna, inserted at middle of third joint. Vertex shining green. Postocular cilia except upper five white. Thoracic dorsum green, more or less shining, towards front and sides brassy. Abdomen shining green, sparsely silvery dusted above, becoming thickly at sides and below, cupreous towards tip. Hypopygium piceous, dusted, greenish towards base, shining on inner surface. Lamellae elongate,



FIG. 18.—*D. pernix*: male antenna and tip of fore tarsus.

light yellow, narrowly margined with black, fringed with dark hairs, inner and apical angle prolonged into several long filaments. Pleurae glaucous, in different parts green, cupreous or piceous, according to angle of vision. Middle and hind coxae piceous, glaucous. Fore coxae yellow, piceous and dusted basally on posterior face; front surface besides the strong apical bristles with fine dark hairs which are supplanted by lighter ones on proximal portion. Legs yellow except apex of hind tibiae, hind tarsi, and last two joints of front tarsi. The middle and front tarsi increase in density of color from tip of first joint. Hind femora not ciliated, with a subterminal bristle. Hind tibiae not glabrous inwardly. Front tarsi slender, as are the tibiae, nearly twice the length of the tibiae; first joint longest, a little shorter than two following; second and third subequal, fourth shortest, fourth and fifth together about equal to third; fourth and fifth joints flattened. Empodia distinct, yellowish. Wings long, hyaline; costa with a small tubercle at juncture of first vein; third vein converging towards fourth; bend in fourth vein slight; at tip of fifth vein a broad, shallow sinus; anal portion moderately prominent. Tegulae and halteres yellow, the former with long black cilia.

One male taken by Mr. Clermont Livingston at Corfield, Vancouver Island, May 21, 1896.

Though closely related to *discifer*, it appears quite distinct. The more evident points of difference are these:

Pernix: First antennal joint not red beneath; arista inserted near middle of third joint of antenna; numerous dark hairs on anterior face of fore coxae; tip of hind tibiae evidently black for some distance; fourth tarsal joint flattened, black; wings not evidently narrowed at base.

Discifer: First antennal joint reddish on under side; arista beyond middle of third antennal joint; front coxae with white hairs (dark hairs on inner side of female, only); hind tibiae dark at only extreme tip and less on outer side; fifth tarsal joint only black; wings rather narrowed towards base.

The proportion of the tarsi to the tibiae is also different, as is also the comparative length of the tarsal joints.

Dolichopus pantomimus, sp. nov.

Green; face narrow, light brown; antennae black with simple arista; cilia of inferior orbit pale; cilia of tegulae black; feet yellow, including front coxae and excepting tip of hind tibiae and tarsi, not ornamented in the male excepting femoral brush; fourth vein not broken.

Male. Length 4 mm., of wing 3 mm. Bright metallic green, somewhat brassy. Proboscis piceous, palpi ferruginous at tip with few dark hairs. Face very narrow, with eyes almost contiguous at middle, thickly overlaid with ferruginous dust, shining. Antennae black, sericeous, not noticeably bristly; second joint closely applied to the third; first joint equal to second on inner side; third joint long, pointed, equal to first two together. Arista finely pubescent, arising from middle of upper surface of second and third joints taken together. Vertex green, shining. Infraocular cilia white. Thorax with dorsum bright green, cupreous anterior to wing insertion, dusted in front; with an indication of two brown median longitudinal lines in front. Abdomen dorsally bright green, cupreous tinged; the posterior margins of segments blackened. Hypopygium wholly piceous, somewhat shining, and finely sericeous. Lamellae in length equal to antennae, white translucent, with a jagged, moderately wide black apical border, and closely fringed with black hairs at tip. Pleurae, sides of abdomen, and base of posterior fore coxae dark green, glaucous. Fore coxae wholly yellow, rather sparsely beset with pale hair, besides the apical bristles. Legs plain, yellow; hind femora with an ante-apical bristle and ciliated below with not long yellow hairs; hind tibiae stouter than the others, and with a long glabrous streak on hind surface, black at tip for one-seventh its length; hind tarsi entirely black, anterior pairs darker towards tip, but not black. Empodia very small, silvery. Wings narrow, tinged somewhat dark gray; costa, at tip of first vein, with an evident knot; fourth longitudinal vein not broken; hind margin entire at tip of fifth vein; anal angle rather strong.



FIG. 19. — *D. pantomimus*:
male antenna and
lamella.

A single male from New Bedford, Mass., collected May 30, by Dr. Garry deN. Hough.

Related to Loew's *melanocerus*, but differs in the smaller size, color of the hairs of the fore coxae, which are not black at base, anterior tarsi not black, and the narrowed darker face.

Dolichopus renidescens, sp. nov.

Green; shining; face broad, light brown, antennae black, with a plain arista; vertex violet; cilia of inferior orbit white, of tegulae black; legs yellow, except tips of the tarsi and hind legs from outer portion of hind tibiae, not ornamented except the ciliation of hind femora; fourth vein not broken.

Length 4.5–5 mm., of wing the same. Bright green, shining, darker on thoracic dorsum, almost bluish. Proboscis piceous, palpi brunneous. Face

broad. Antennae dull black, sericeous, short, with slender, dark, sericeous arista, once and a half the length of the antenna; third joint a little shorter than the first two together, broadly oval, rounded but obtusely pointed at apex; second joint with circlet of hairs. Front violet. Upper seven of postocular cilia black, rest pale yellow. Thoracic dorsum bluish-green, brilliantly shining except for indications of longitudinal dusted rows; scutellum and ante-scutellar region purer green. Abdomen shining green, with brassy tinge, lightly dusted. Pleurae glaucous on a green foundation. Middle and hind coxae, except tip, and extreme base of fore coxae of same color as pleurae. Front coxae with black pubescence on anterior face. Legs largely yellow, the hind femora with two ante-apical bristles; fore and middle legs dark from tip of first tarsal joint; hind tarsi black, hind tibiae infuscated at tip. Wings hyaline, normal, a slight sinus at tip of fifth longitudinal. Tegulae yellow

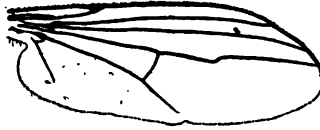


FIG. 20. — *D. venidocens*: male wing and lamella.



with rather long black cilia. Halteres yellow.

Male. Face ferruginous. Hypopygium piceous with brassy green tinge; sericeous below, shining inwardly; internal appendages yellow. Lamellae clavate, broad, white translucent, rather broadly margined with black at extremity, apex jagged and fringed with rather long, slender, nearly straight, black hairs. Hind tibiae with a long, narrow glabrous streak, more evident near tip, on hind face. Anal angle of wing full; costa thickened at junction with humeral vein.

Female. Face with gray dust. First antennal joint a little longer than in male. Hind tibiae not glabrous, the apical infuscation not evident. Anal angle of wing rounded; costa not thickened.

Two males and one female from North Park, Colorado, collected at an altitude of over 9000 feet during July.

The shorter antennae, broader face, violet front, more extended margination of hypopygial lamellae, and the closer ciliation with brown hairs of the hind femora which possess two ante-apical bristles, distinguish this species from *melanocerus* Loew.

Dolichopus apheles, sp. nov.

Green; face ochraceous; antennae black, with a simple arista; infra-orbital cilia white; tegular cilia black; feet plain, yellow, except tips of hind femora and tibiae black; hind tarsi black; fore coxae yellow with dark hairs; fourth longitudinal vein not broken.

Male. Length 5 mm., of wing 4 mm. Not so brightly colored as in most species, largely green. Proboscis piceous, palpi roseous yellow. Face

ochraceous. Antennae sericeous, black, except underside of first joint, which is indistinctly reddish, very like those of a female *Hygroceleuthus*; first joint longer than second, short, hairy above; second with a crown of black bristles; third short, deep, subtriangular. Arista sericeous. Vertex blue green in certain lights, violet in others, somewhat shining. Infraocular cilia pale; six of the supraocular cilia black. Thorax dull, bluish on dorsum; posterior declivity and scutellum shining green. Abdomen shining green dorsally, cupreous toward apex, transverse margins of segments piceous. Hypopygium piceous with greenish tint, shining, and not sericeous on inner face; lamellae rounded, rather short, white translucent, with a narrow, black, apical border, jagged and fringed with black hairs. Pleurae glaucous, as are the middle and hind coxae, except tips. Front coxae yellow with a basal glaucous-piceous spot on the outer side; front surface with a coating of short black hairs, besides apical bristles. Legs yellow, entirely unornamented; the darker places are: hind tarsi and outer fourth of hind tibiae black, tip of hind femora more evidently on upper surface black; the infuscation of fore and middle tarsi begins at middle of first joint. Hind femora with a single ante-apical bristle and not ciliated beneath; hind tibiae with no evidently glabrous space. Wings normal, rather dusky anteriorly; without costal thickening at tip of first vein; fourth vein unbroken, beyond bend gradually converging with third, but almost subparallel with it; no indentation in posterior margin; anal angle full. Tegulae and halteres yellow, tegular cilia black, rather short and stout.

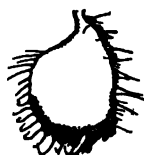


FIG. 21. — *D. aphotes*: male lamella.

One male collected by Dr. Wm. M. Wheeler near Milwaukee, Wis., June 28, 1895.

This unique species is allied nearest to those species grouped about *melanocerus* Loew and *incisuralis* Loew.

The addition of the last four species has necessitated the following modification of Divisions 52 to 56 of Professor Aldrich's table.¹

52. Front legs of male ornamented	2
Front legs plain	3
2. Fourth joint of fore tarsi of male not flat	<i>discifer</i> Stan.
Fourth joint of fore tarsi of male flat, black	<i>pernix</i> sp. nov.
3. Antennae wholly black; hind femora of male ciliated	4
First antennal joint lighter below	6
4. Front coxae with light hairs	<i>phantomimus</i> sp. nov.
Front coxae with dark hairs in front	5

¹ *Kan. Univ. Quart.* Vol. ii, No. 1, p. 5.

5. Face rather narrow; front green *melanocerus* Loew
 Face broad; front violet *renidescens* sp. nov.
 6. Femora of hind legs of male ciliated, not blackened 7
 Male hind femora not ciliated, black at tip *apheles* sp. nov.
 7. Front coxae with black pubescence 8
 Front coxae with white pubescence *platyprosopus* Loew
 8. Bristles of hind tibiae long *setosus* Loew
 Bristles of hind tibiae normal *incisuralis* Loew
56. *praeustus*, etc.

Dolichopus amphericus, sp. nov.

Light green; antennae yellow, except third joint and tip of second; fore tarsi ornamented; femora yellow; postocular cilia pale below; tegular cilia black, hind tibiae not black at tip.

Male. Length 6.5–7 mm., of wing 5.5–6 mm. Light coppery green with much white dust. Proboscis piceous, palpi testaceous. Face of medium width, about four times as long as broad, thickly covered with brilliant yellow dust. Front shining green. Antennae rather elongate; first joint yellow, with many short black hairs above; second joint yellow at base, becoming black at apex; third joint black, sericeous, obtusely

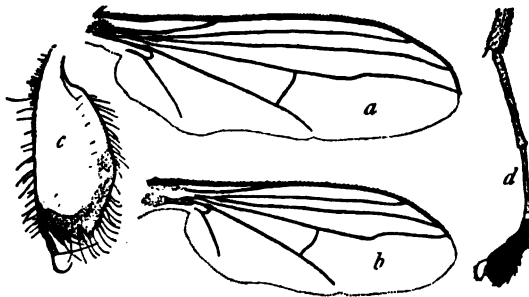


FIG. 22.—*a*, *D. amphericus*, male wing; *b*, *D. coloradensis*, male wing;
c, *D. amphericus*, lamella; *d*, *D. amphericus*, male fore tarsus.

pointed at apex. Arista less than twice as long as antenna, very distinctly pubescent. Postocular cilia black above and light yellow on lower three-fourths. Thorax light green, coppery on the disc; slightly opaque by the presence of light yellow dust. Dorsally there is a deep coppery longitudinal stripe. Abdomen shining green, white dusted. The white dust is so thick as to obscure the ground color on the lower part of the sides. Incisures coppery. Hypopygium black, shining, except at base, where it is white dusted. Near the base bearing a large patch of black hair. Internal appendages ferruginous; lamellae very pale yellow, with a wide, sharp border of black at apex, where they are bristly and deeply toothed. Outer tooth bearing at its tip a strong, curved bristle. Pleurae greenish-black,

white dusted. Fore coxae yellow with white pubescence in front, at apex and inwardly with black hairs. Middle and hind coxae of same color as the pleurae, yellow only at extreme tip. The middle pair with white hairs in front. Legs yellow. Fore tarsi ornamented; the first two joints long and slender, first about once and a half the length of the second; third less than one-half the length of the first, much enlarged at apex, where it is infuscated; fourth joint small, shorter than the third, flattened, velvety black; fifth oval, about one-half as long as the first, broadly compressed, deep black and fringed on anterior edge with black hairs; empodia silvery white. Middle tarsi infuscated from tip of first joint. Hind femora not ciliated; hind tibiae wholly yellow with a dorsal, apical, glabrous stripe; hind tarsi wholly black. Tegulae and halteres yellow; tegular cilia black. Wings narrow, nearly hyaline, slightly brownish in front; costa with no noticeable swelling; fourth vein not broken; distinctly lobed at tip of sixth vein.

Female. Length 5.5–6.5 mm., of wing 6.25–6.75 mm. Face yellowish-gray. Front tarsi plain, infuscated from tip of first joint, the second and third joints lighter at base, giving the tarsus a somewhat banded appearance. Wings darker and longer than in the male; only a faint indication of the preanal lobe.

Two males and three females from Price County, Wis.; collected by Dr. Wm. M. Wheeler.

This species resembles *coloradensis* Aldrich, from which it differs by the larger size, bright yellow face, lighter antennae, brownish wings, and white hair on front face of anterior coxae.

Together with *flagellitenens* Wheeler, *amphericus* possesses greatly enlarged metapleurae which give a winged appearance to the first abdominal segment. The posterior portion of the metapleurae is dull black and pubescent.

The following localities are those of species in the collection of Dr. Wm. M. Wheeler:

Group *Hygroceleuthus*.

<i>latipes</i> Lw.	Wisconsin, Illinois.	fornia, Washington, Wyoming,
var. <i>cognatus</i> ,	Illinois, Massachusetts.	Idaho.
<i>Aldrichii</i> Wheeler.	Idaho, Wyoming, Colorado.	<i>consanguineus</i> Wheeler. California.
<i>plumipes</i> Scop.	Colorado. Vancouver.	var. <i>propinquus</i> . Vancouver Island.
<i>Wheelerii</i> M. et B.	Massachusetts,	<i>afflictus</i> O. S. Arizona, California, Washington.
<i>amnicola</i> M. et B.	Colorado.	<i>ciliatus</i> Ald. Wvoming, South Dakota.
<i>crenatus</i> O. S.	Vancouver, Cali-	<i>idahoensis</i> Ald. Idaho.

Group *Dolichopus*:

- partitus* M. et B. Colorado.
paluster M. et B. California.
laticornis Lw. Wisconsin, Wyoming.
intentus M. et B. Illinois.
incongruus Wheeler. Wisconsin.
gratus Lw. Illinois, Wisconsin.
calcaratus Ald. Massachusetts.
detersus Lw. Illinois, Wisconsin.
myosota O. S. California.
calainus M. et B. Illinois.
acuminatus Lw. Illinois, Wisconsin.
ovatus Lw. Wisconsin.
enigma M. et B. Colorado.
setifer Lw. Wisconsin, Massachusetts.
albiciliatus Lw. Massachusetts, Illinois, Wisconsin.
agronomus M. et B. Massachusetts.
xanthocnemus Lw. Vancouver Island.
pachycnemus Lw. Massachusetts.
longimanus Lw. Wisconsin, Massachusetts.
albicoxa Ald. Massachusetts, Illinois.
brevimanus Lw. Massachusetts, New Hampshire.
socius Lw. Massachusetts, New Jersey, Wisconsin.
palaestricus Lw. Illinois, New Hampshire.
splendidus Lw. Ontario, Michigan, Illinois.
splendidulus Lw. Illinois, New Hampshire.
batillifer Lw. Massachusetts.
tonsus Lw. Massachusetts.
tener Lw. Wisconsin.
variabilis Lw. Illinois, Wisconsin.
lutiepennis Lw. Vancouver Island.
bifRACTUS Lw. Massachusetts, Illinois, Nebraska.
obcordatus Ald. Wyoming, Idaho.
- ramifer* Lw. Illinois, Texas, Wyoming.
vittatus Lw. Illinois, Wisconsin.
cuprinus Wied. Illinois, Wisconsin, Wyoming.
longipennis Lw. Vancouver Island.
flagellitenens Wheeler. Illinois, Wisconsin.
comatus Lw. Massachusetts, Illinois, Wisconsin.
pernix M. et B. Vancouver Island.
melanocerus Lw. Massachusetts.
pantomimus M. et B. Massachusetts.
renidescens M. et B. Colorado.
apheles M. et B. Wisconsin.
setosus Lw. Massachusetts, Vancouver Island.
gracilis Ald. Wisconsin.
angustatus Ald. Massachusetts.
lobatus Lw. Illinois, Wisconsin, Michigan.
coloradensis Ald. Colorado.
amphericus M. et B. Wisconsin.
Henshawi Wheeler. Massachusetts.
marginatus Ald. Massachusetts, New Jersey.
scoparius Lw. Massachusetts, Illinois, Wisconsin.
canaliculatus Thomson. California.
duplicatus Ald. Idaho.
Coquilletti Ald. Idaho, Vancouver Island.
tenuipes Ald. Idaho, California.
occidentalis Ald. Idaho, Vancouver Island.
scapularis Lw. Wisconsin.
germanus Wheeler. Wisconsin, Wyoming.
grandis Ald. California.
sexarticulatus Lw. Illinois, Louisiana.
Willistonii Ald. Kansas.
terminalis Lw. Wisconsin.
sarotes Lw. Wisconsin.

ON THE ORIGIN OF THE SPERM-BLASTOPHORE OF SOME AQUATIC OLIGOCHAETA.

SHINKISHI HATAI.

THE first investigator to discuss the origin of the sperm-blastophore in Oligochaeta was Bloomfield.¹ His work was done mostly on living material, although he supplemented it to some extent by preparations mounted in glycerine. He studied the external features only. Later, Calkins² published a detailed account on the same subject, his views being opposed to those of Bloomfield. Both of these writers made their observations upon *Lumbricus terrestris*. A more complete statement of their respective views will be given on a later page.

In the Limicolae the origin of the sperm-blastophore has not yet been studied, although some work on the structure of *Limnodrilus Gotoi*³ and *Vermiculus limosus*⁴ of this group has been published recently. These species are common in Japan. The material used in the present study was fixed in Perenyi's fluid and corrosive sublimate. The stains used were Kleinenberg's haematoxylin, Rawitz's haematoxylin, and borax carmine.

The present article deals only with the formation of the sperm-blastophore. The various stages in its development may be described advantageously in the following order:

1. *Spermatogonia*.—A section of the testis (Fig. 1) shows three stages in the maturation of the spermatogonia, namely: (a) The cells at and near the proximal end of the testis are

¹ Bloomfield, E., "On the Development of the Spermatozoa. 1. *Lumbricus*," *Quart. Journ. Micr. Sci.* Vol. xx. 1880.

² Calkins, G. N., "The Spermatogenesis of *Lumbricus*," *Journ. of Morph.* Vol. xi. 1895.

³ Hatai, S., "On *Limnodrilus Gotoi* (n. sp.)," *Annotationes Zoologicae Japonesis*. Vol. iii, Part i, 1899.

⁴ Hatai, S., "On *Vermiculus limosus*," *Annotationes Zoologicae Japonesis*. Vol. ii, Part iv, 1896.

somewhat polygonal, firmly connected, and possess comparatively small nuclei. (b) The cells of the central part are larger than the former, becoming gradually spheroid and more loosely connected; the nuclei are larger, and the peritoneal membrane of the testis disappears in this region. (c) At the free ends of the testis the cells present completely spherical forms, and are so loosely connected that they may be easily detached. The spermatogonia have conspicuous nuclei. Each fully matured spermatogonium has one large nucleus, and is not

multinucleate, as in *Lumbricus* (Calkins).

2. *Spermatocyte* (Figs. 2, 3). — The primordial germ-cells,

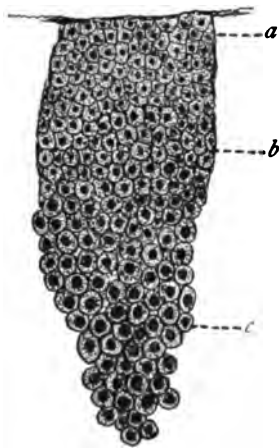


FIG. 1.

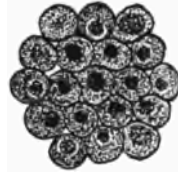


FIG. 2.

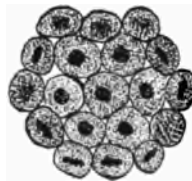


FIG. 3.

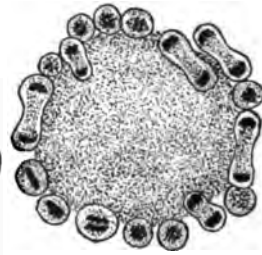


FIG. 4.

or spermatogonia, when fully matured drop from the testis; sometimes one or two, but usually many at the same time, afterwards swelling up gradually. When they are first detached from the testis, no changes are noticeable, but with the beginning of cell division the following nuclear changes are observed: The nucleus of each cell in the outer layer moves inward toward the center of the cluster, as shown in Fig. 2. Each nuclear membrane disappears, while the chromosomes become distributed evenly throughout the nucleus. Soon the chromosomes collect in the equatorial plane (Fig. 3) and undergo division by the usual method of karyokinesis (Fig. 4). The spermatid arises after two or more such divisions. The cells at the central part show no signs of change, but remain in the resting stage.

Usually, after the peripheral cells have divided, the central cells become granular and appear homogeneous; the nuclei and cell membranes can no longer be distinguished (Fig. 5). In other words, the central cells degenerate at this period and become transformed into a cushion for the spermatozoa. This cushion is called the "sperm-blastophore" by Bloomfield.



FIG. 5.

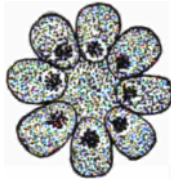


FIG. 6.

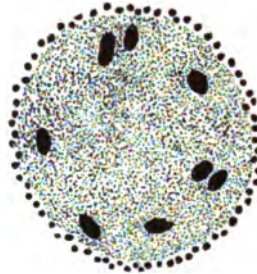


FIG. 7.

(Occasionally this transformation of the central cells occurs previous to the division of the peripheral cells, Fig. 6). The daughter-cells produced by the several divisions of the spermatocyte become half the size of the mother-cells, and retain this size throughout the stages of formation of the spermatozoan. These half-sized cells are the spermatids.

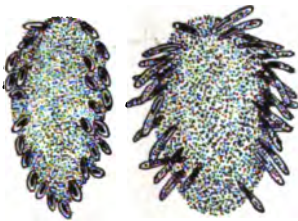


FIG. 8.



FIG. 9.



FIG. 10.



FIG. 11.

3. *Spermatids*.—The spermatids encircle the blastophore (Fig. 5), which changes to a spherical form and becomes more conspicuous than in the former stage. The spermatids now undergo repeated cell division, producing an enormous number of new spermatids (Fig. 7), which gradually elongate (Figs. 8, 9), and finally become tailed spermatozoa (Figs. 10, 11). The

tails of the spermatozoa all turn in one direction, as depicted in Figs. 10, 11. While these changes of the spermatids are in progress, the blastophore itself changes its form from spheroid to oblong and sometimes becomes spindle-shaped (Fig. 12).

As shown above, the central cells of the original cluster degenerate to form the sperm-blastophore, which appears as a homogeneous substance. It should be here stated that cases have been observed in which several nuclei were scattered through the homogeneous substance of the blastophore, but rarely a complete cell, as shown in Fig. 13. The question arises as to the origin of these nuclei or cells. It seems rea-

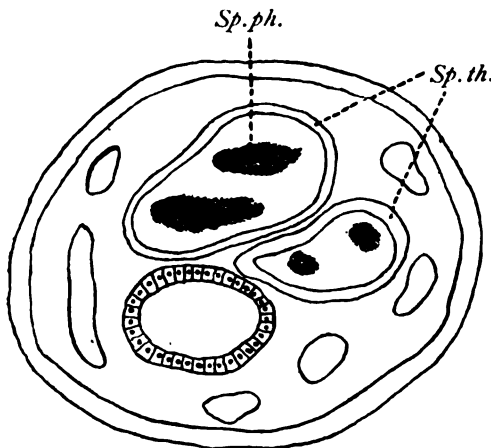


FIG. 12.

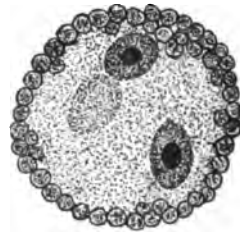


FIG. 13.

sonable to suppose that they are simply belated portions which have not yet been transformed into the homogene-

ous mass. Fig. 7 shows a condition in which only the nuclei remain. At no time during the formation of the blastophore has a proliferation of the central cells been observed.

The spermatophore becomes slightly modified in passing from the sperm-sac to the spermatheca, as shown in Figs. 12, 14, 15. It is to be noticed that the tails of the spermatozoa turn spirally; this being a secondarily acquired character occasioned by its passage through the sperm-duct. Following these changes, the sperm-blastophore becomes more or less spindle-shaped; it also decreases in size. In the cross-section of a spermatophore a central canal is generally to be seen, as represented in Fig. 15. The blastophores are at first com-

paratively large (Figs. 8-11); they gradually decrease, and finally disappear, as shown in Fig. 15. It would appear, therefore, that the blastophore is produced by the degeneration of the central cells, and that it not only acts as a cushion, affording a means for conveying the spermatozoa, but also serves as nourishment for them.

Bloomfield's principal results, as briefly summarized by Calkins, are as follows:

"1. The early germ-cell is not entirely used in the formation of spermatozoa; a central part remains passive, and serves to carry the developing spermatid cells. This central part is called the sperm-blastophore, and may or may not be nucleated.

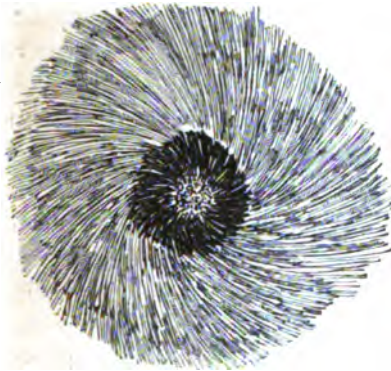


FIG. 14.

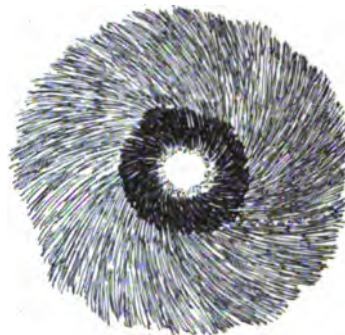


FIG. 15.

2. The sperm-blastophores increase by division while in the testis, and disappear, probably by atrophy, after the spermatozoa leave it.

3. The blastophore corresponds to the nucleated supporting cells (Sertoli's cells) of the frog and salamander.

4. The large nucleus of the early sperm-cell divides many times to form secondary nuclei, which stand out around the central mass, or blastophore, of the generating spheroid with very little protoplasm clothing them. These nuclei become the rod-like heads of the spermatozoa.

5. The protoplasm collects in a small cup, or knob-like mass, at the distal end of the developing cell, and from this grows out the long vibratile tail of the spermatozoan. (This 'mass' must be the archoplasm of the spermatid.)"

Calkins summarized his own results as follows :

"1. A multinucleated cell is formed in the testis; this represents a group of the earliest spermatogenic cells or spermatogonia. Each spermatogonium gives rise to several spermatozoa.

2. The nuclei arrange themselves around the periphery of the multinucleate cell; cytoplasmic cleavages then ensue between the nuclei, as in the centrolecithal egg. The cleavage grooves deepen until the nuclei are separated from the central mass of cytoplasm by mere filaments.

3. The residual mass of cytoplasm thus formed (the blastophore) is not nucleated, and cannot be compared with a Sertoli's cell in function, form, or mode of origin. It finally disappears. The blastophore furnishes perhaps the chief source of food supply for the parasites — monocysts — which live in the seminal vesicles. A possible explanation of the function of the blastophore is that of superfluous nutritive cytoplasm, the vital protoplasm having gathered around the nuclei."

Thus it will be seen that Bloomfield and Calkins hold very different views regarding the blastophore. The former considers it as having a nutritive or feeding function. It carries developed spermatozoa, and is to be regarded as the homologue of Sertoli's cell. The latter maintains that the blastophore is merely an excess of cytoplasm and not a true cell; therefore it cannot be homologous with the Sertoli cell.

The question as to the structure and function of the blastophore in *Lumbricus* can be decided only when we learn its true origin. In the present work on the two new species of *Limicolae*, it appears that the blastophore originates through the degeneration of certain of the primordial germ-cells which lie at the centers of the clusters of spermatogonia given off from the testes.

It serves not only for carrying developed spermatozoa, but for their nourishment as well. Thus it arises from *definite cells*, and, as Bloomfield has suggested, may be compared to the *Sertoli's cell*.

HULL ANATOMICAL LABORATORY,
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PECULIAR TRACHEAL DILATATIONS IN BITTACOMORPHA CLAVIPES FABR.¹

CHARLES THOMAS BRUES.

BITTACOMORPHA is a member of a very aberrant group of *Tipulidae*. In connection with two other genera it has been separated from the *Tipulidae* and considered as a distinct family. Of the genus *Bittacomorpha* only two species are known, both from North America. The species upon which these remarks are based is the commoner and more widely distributed form. It occurs from the New England states westward to the Pacific coast and has been taken as far south as Florida by Osten Sacken. In the northern states it is double brooded, and the imagines are seen during May and September, although much more commonly in the spring. The other species (*Sackenii*), which was described by Von Roeder in 1890,² is much more limited in its distribution and is recorded only from Nevada.

The common species (Fig. 3) is of the very slender form so characteristic of the *Tipulidae*. Its appearance is remarkable, however, on account of the peculiar black and white banding and the great inflation of the metatarsi of all the legs. The preparatory stages of a European species of the closely allied genus *Ptychoptera* have long been known, but it was only very recently that the larva and pupa of *Bittacomorpha clavipes* were discovered and figured by Hart.³ The larva, like that of *Ptychoptera*, is aquatic, living among the submerged brushwood and sticks, which it resembles in color and external appearance. It is in this instar that we find the first peculiar modification of the tracheal system. The larva is furnished with an elon-

¹ *Contributions from the Zoölogical Laboratory of the University of Texas*, under the direction of Wm. M. Wheeler, No. 3.

² *Wiener Ent. Zeitung*. Heft 8, p. 230.

³ *Bull. Illinois State Lab. Nat. Hist.* Vol. iv, p. 193.

gated breathing tube, produced by the excessive lengthening of the posterior end of the abdomen (Fig. 1). This formation is not peculiar to this species, as it occurs elsewhere, even in insects in nowise related, as in *Eristalis* among the *Syrphidae*. In the pupa we find a respiratory tube present, but in this instar its insertion is exactly reversed; it proceeds from the head. Although only one tube is functionally developed, it is one of a pair which has lengthened at the expense of its fellow (Fig. 2). Moreover, it is not always the same tube which is developed. Hart mentions that twenty-seven pupae had the right tube elongated as against three in which the left tube functioned. In one anomalous case both were developed, but unequally, their combined length being equal to that of the long one in normal pupae. This unequal length of the tubes is characteristic also of *Ptychoptera*.

Up to the present time it has not been known that the imago also possesses a remarkable modification of the tracheal system. In this stage, however, it is to be found in the legs.

In both sexes the metatarsi are very much enlarged and quite conspicuous on account of their great color contrast. The second and third tarsal joints are also somewhat enlarged, but not nearly to so great an extent.

In order to study the tracheal system of the legs they were decolorized in chlorine water and mounted whole or split into halves. Some specimens were treated also with potassium hydroxide, which successfully separated the delicate tracheae from the integument. Legs were also sectioned in paraffin to show the disposition of the internal parts.

In the basal part of the legs the tracheal tube is of the ordinary form and size. It begins to enlarge just before the middle of the femur, and before it has reached the tip is equal to seven-eighths the diameter of the femur. At this point the taenidia extend entirely around the tube, although faint in some places. The whole tibia is completely filled up by the trachea, which is striated on each side for only about one-fifteenth of its circumference. In the enlarged metatarsus the trachea is enormously distended and almost completely fills the cavity of this joint as well as that of the second and third

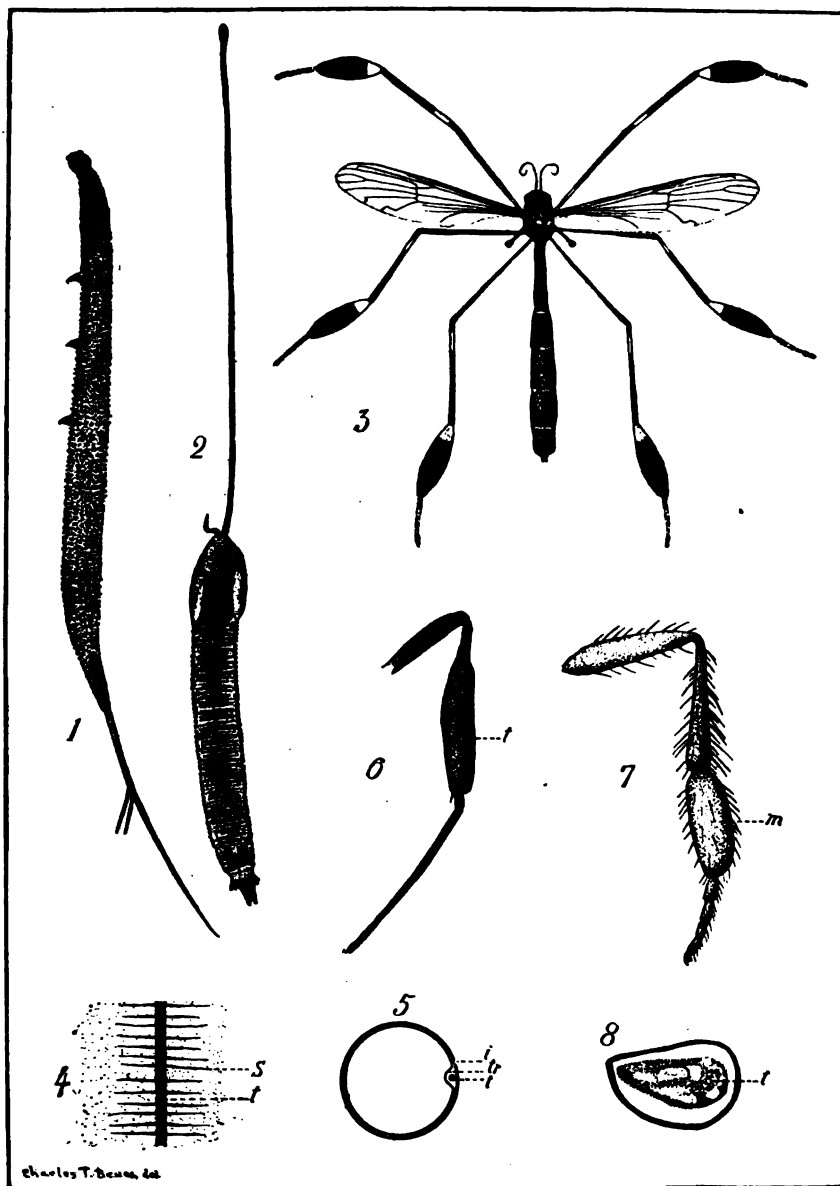


Fig. 1. Larva of *Bittacomorpha clavipes* (after Hart).—Fig. 2. Pupa of *Bittacomorpha clavipes* (after Hart).—Fig. 3. *Bittacomorpha clavipes*, in the position which it assumes when flying.—Fig. 4. Portion of tracheal wall of metatarsus of *Bittacomorpha*, showing position of tendon; *s*, taenidial striation; *t*, tendon.—Fig. 5. Diagrammatic cross-section of metatarsus of *Bittacomorpha*; *t*, tendon; *i*, chitin integument; *tr*, tracheal wall.—Fig. 6. Hind leg of *Pelocinus polyturator*, ♀; *t*, tibia.—Fig. 7. Foreleg of *Hilara trivittata*, ♂; *m*, metatarsus.—Fig. 8. Cross-section of metatarsus of *Hilara trivittata*, ♂; *t*, trachea.

joints of the tarsus. The wall of the trachea lies closely applied to the exoskeleton of the metatarsus, except for a very short distance on one side of the leg, where it is semicircularly bent inward to leave space for the claw tendon which lies in the tubular space thus formed (Fig. 5). Here as in the tibia the taenidia are visible on only about one-fifteenth of the circumference on each side.

The walls of the trachea are here not entirely destitute of taenidia, as is the case with the air vesicles which appear in the body cavities of many active insects. The striations have remained on that part of the tracheal wall which encloses the tendon (Fig. 4). At the point where the tendon passes, the taenidia are thickened and quite robust, but on each side they gradually become weak and fade out entirely. An exactly symmetrical formation of the taenidia is present on the side opposite to the tendon. It is evident that the thickenings on the tendon side may have been retained in order to strengthen the tube at this point, but there is apparently no reason for the anomalous thickening on the opposite side. In the second and third joints the taenidia lengthen until they extend over one-seventh of the circumference. The trachea seems to stop suddenly here, as I have been unable to trace it further.

There are few insects presenting similar enlargements of the leg joints, if we except those forms such as jumping *Orthoptera* and *Chrysomelidae*, where the increase in size is evidently for the accommodation of the larger muscles. Graber and Lubbock mention enlargements of the trachea in the tibiae of *Orthoptera*, ants and *Termitidae*, serving as auditory or chordotonal organs. In this case the adaptation is very extraordinary, but the dilatation of the trachea is not comparable to that of *Bittacomorpha* in extent. *Bittacomorpha* presents the only case known to me of a considerable tracheal dilatation occurring in the insect leg. In the males of many *Empididae*, and notably species of *Hilara*, e.g., *Hilara trivittata* Lw., the metatarsus of the front leg is greatly enlarged (Fig. 7), but here the cavity is occupied in great part by muscular tissue, the trachea being very slender (Fig. 8). In this species there seems to be no trachea beyond the end of the metatarsus.

In the legs of normal *Tipulidae* (*Pachyrrhina* sp.? and several other species) the trachea occupies a considerable space only in the femur and tibia, where it fills up from one-fourth to one-sixteenth of the cavity. In the tarsus the tracheal tube is very delicate or obsolete.

The female of the peculiar parasitic hymenopteron, *Pelecinus polyturator* Drury, presents an external appearance similar to that of *Bittacomorpha* in the enlarged hind tibia (Fig. 6). Here, however, the chitin of the external wall is thick and heavy, and the trachea is robust, strongly striated, but not at all dilated.

It is the rule in insects, wherever a tracheal dilation occurs, that the taenidia become obsolete, but the thinning of the tracheal wall can nearly always be regarded as a modification for the purpose of offering less resistance to osmosis. This is illustrated by the air vesicles in the bodies of insects, which are generally considered to be reservoirs for storing air to be used during extended muscular exertion. The presence of these immense vesicles in the metatarsi cannot be explained on the same principles, for it is impossible that they should serve as reservoirs for air to be used in respiration, on account of their distance from the body of the insect. It is more probable that they may bear some relation to the insect's method of locomotion. When flying, *Bittacomorpha* uses the wings scarcely at all, relying in great measure upon wind currents for transportation. The legs are exceedingly light, as the exoskeleton is thin and delicate, and encloses practically no tissue which can serve to increase their weight. As they expose a large surface, they offer great resistance to the air without adding appreciably to the insect's weight.

Drifting along thus, their extremely slender bodies and white banded appendages give them a most peculiar, intangible appearance, which is heightened by their extremely slow motion.

When examined in the cabinet, the conspicuous white and black banding of *Bittacomorpha* seems to point toward a case of warning coloration. When they are seen against their natural background, however, all these brilliant contrasts fade away into a perfectly neutral color which causes them to resem-

ble a spider's web or thistle seed drifting along. Indeed, when I first saw them it was hard to believe that they were really alive. *Bittacomorpha* seems to have developed its protective coloration along the same lines as the zebra among the vertebrates; it is rendered invisible not only by the fragmentary distribution of color, but by the neutral gray color produced by the visual blending of the black and white.

From the description of *Bittacomorpha Sackenii* by Von Roeder, it seems that this species does not possess any dilatation of the metatarsi. It would be very interesting to see if there is any abnormal development of the tracheal system in this species, but unfortunately I have been unable to procure specimens. The specimens examined were given to me by Dr. Wm. M. Wheeler, to whom I feel greatly indebted for many kind suggestions.

LAMPREYS IN CAPTIVITY.

ALBERT M. REESE.

HAVING had living lampreys of various ages under observation in the biological laboratory of Johns Hopkins University, I present the following facts as to the ability of these animals to live in a very limited space.

I received, about the middle of May, from Ithaca, N. Y., two lots of lamprey eggs, about six dozen eggs in each lot. They were shipped by express and must have been on the road about twenty to twenty-four hours. They had been shoveled out of the "nest," with about 2 l. of gravel, and put into two tin buckets of 8 l. capacity. The space in the buckets above the gravel was filled with water, and in one of the buckets were some three dozen larval lampreys ranging from 2 cm. to 12 cm. in length. None of these eggs developed, although they were put into running water as soon as they reached the laboratory.

My experience with the small larvae (about 5 mm. in length) was more successful. I obtained one hundred or more of these from a stream at Ithaca, and brought them to Baltimore in two glass jars of 3-4 l. capacity each. A small quantity of gravel was placed in the bottom of each jar for the larvae to bury themselves in, and the water was kept cool by partially emptying the jars from time to time, and refilling them with ice water from the coolers on the train. The journey lasted for about eighteen hours, and all the larvae, except three or four, reached the laboratory in good condition.

The small amount of sediment in the city water proving disastrous to the welfare of the larvae, clear spring water was obtained every few days, and this was kept cool by allowing the jars to stand in larger vessels of running water. Even with this arrangement the deaths averaged one per day, and about the first of August the remaining larvae were killed and

preserved, after having, with difficulty, been kept alive for six weeks.

The older larvae which were received, as has been said, in one of the buckets containing eggs, proved to be very hardy, and five or six of them were kept in a 12 l. aquarium for six months or more without the least difficulty. At the end of this time they were killed, two of them having shortly before (October 20) transformed into the adult *Petromyzon branchialis*. During their entire captivity they remained completely buried in the sand in the aquarium. A small stream of water was kept running through the aquarium, though a *constant* change of water was not necessary.

In the early part of April, I brought from the herring fisheries at Port Deposit, Md., five large sea lampreys (*P. marinus*) in two tin buckets, each bucket of about 50 l. capacity. Being nearly a meter in length and about 12 cm. in circumference, the five lampreys were rather crowded in the two buckets, and only four of them survived the three-hour journey to the laboratory. They were put into an aquarium (1.5 m. $\times$.8 m. $\times$ 12 cm.) of running water, where they lived comfortably for several weeks, until by accident the wire screen was left off the aquarium, and three of them escaped and were found dead upon the floor. On June 22 the remaining lamprey was killed. It proved to be a female, and 250 cc. of ripe ova were "stripped" from her with ease. Had one of the males been kept alive it seems probable that artificial fertilization could easily have been accomplished.

To sum up, then, it seems (1) that the very small larvae are very delicate and hard to keep in confinement; (2) that the large larvae are unusually hardy; and (3) that the adults are able to live in captivity moderately well.

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BIOLOGICAL BULLETIN.

OUR NORTH-AMERICAN ECHIURIDS.

A CONTRIBUTION TO THE HABITS AND GEOGRAPHICAL RANGE OF THE GROUP.

CHAS. B. WILSON.

ONLY four species of Echiurids, representing three of the five known genera, have been reported from the North American coast up to the present time. These are as follows: *Thalassema mellita* Conn was found at Beaufort, Va., living in empty sand-dollar tests, and its structure and morphology were most admirably worked out (2). *Thalassema viridis* Verrill has been reported from nodules of blue clay at a depth of seventy-seven fathoms off Head Harbor, Campobello Island (16). *Bonellia sulmii* Selenka was dredged from deep water off the coast of Nova Scotia and was described and named in the *Challenger Reports* (12).

The fourth species, *Echiurus chrysacanthophorus* Pourtales, has been reported by several authors from various localities on the New England coast. Each of these four was established as a new species by its discoverer and has been found nowhere else. The *Bonellia* species was based upon a single badly mutilated specimen, *T. viridis*, and the species of *Echiurus* upon a very limited number of specimens, no one of which in the latter species was perfect, leaving *T. mellita* to stand alone beside the well-known and thoroughly studied European forms.

In the present contribution I am able to add American localities for two of the well-known Old World species, and when

the results of the recent Harriman Alaskan Expedition are published another even better known European form will be found among them.

It is hoped also to clear up the cloud of doubt which has hung about the questionable American *Echiurus chrysacanthophorus*, for the reports on this species have contained so many gross errors and conflicting statements, and so little accurate description, that the determination of the exact species has been impossible.

This has been due to a variety of causes, chief among which may be mentioned two. First, it is essentially a shore species, frequenting muddy shallows where the water is too deep or too roily for the shore collector and not deep enough for dredging. Consequently only a limited number of specimens have been obtained.

Again, so far as known, every one of these was so mutilated in the getting as to render a full description impossible. The part most easily injured is the delicate proboscis. This breaks off upon the slightest provocation, and leaves no scar that can be detected even with a good hand lens.

Hence it is difficult to obtain a specimen with the proboscis intact even under favorable circumstances, and absolutely impossible by dredging. It was this denudation of the proboscis with no resultant scar which led the discoverer of the species, Couthouy, to mistake it for a holothurian, and to describe it as *Holothuria chrysacanthophora* in 1838 (3).

The same mistake was made by Gould in 1841 (5), who says: "This is not unlikely to be *H. forcipata* of Fabricius. Several specimens which I have seen were all taken from fishes' stomachs in a mutilated state. Some of the essential characters, therefore, remain yet undetermined. The surface is light colored and appears to be naked, except that there are several long, flexible, sharp-pointed spines about the mouth¹ of a shining golden yellow. One specimen is five or six inches in length."

Pourtales rectified this mistake in 1851 and located it correctly among the Gephyrea, giving it a name which it has since

¹ Really the anus.

borne, *Echiurus chrysacanthophorus* (8). But he also adds: "I have seen but a single specimen of this species. The one I have before me answers very well to the characters assigned to *Echiurus Gaertneri* by Quatrefages. It wants likewise the spoon-shaped appendage," *i.e.*, the proboscis (*ibid.*).

But Quatrefages himself admitted in 1865 that the species *Gaertneri* was based "upon individuals which had been rolled about by the wind," and he adds: "It is very possible that the appendage was broken off" (10). This proved to have been the case, and in 1880 Greef included the species *Gaertneri* as one of the synonyms of *E. Pallasii* Guerin, but he retained the doubtful species *chrysacanthophorus* (7).

A species of *Echiurus* has been dredged by Professor Verrill at various points along the New England coast, and has been reported conditionally as *E. chrysacanthophorus*. But Verrill wrote me in 1895 that in all his dredging (over 1000 localities) he had met with this species "in very few instances, and *never*¹ a perfect specimen." Hence he wisely refrained from attempting any detailed description of it, and from any comparison with foreign species.

Finally Shipley, in 1896 (13), and again in 1899 (14), rejects the species altogether as being inadequately described, and the locality, "North Atlantic," which he assigns to *Echiurus Pallasii*, doubtless signifies the Norwegian and Greenland shores, from which it has been reported by other authors.

Such being the condition of affairs, it seems fitting to describe the species accurately, to determine it definitely, and to add a few observations upon its habits which may be of generic interest. This is rendered possible by the fact that it has been the good fortune of the author to obtain a large number of absolutely perfect specimens and to keep some of them under observation in aquaria for several weeks, while others were successfully preserved,—a by no means easy task.

The material was all obtained at Casco Bay, on the Maine coast, during the summers of 1895-98. It is also hoped that the photographs which accompany these notes may prove of assistance in locating the species.

¹ The underscoring is his.

Habits. — This species lives in the mud near and below ordinary low-water mark. It can be best obtained during the few days of each month when the tides run lowest, at which time it can be dug in the same way as the common clam (*mya*).

It is most abundant in close proximity to mussel beds where the mud is soft and very black with organic matter.

I am aware that this habitat is radically different from that given by Greef and others for *E. Pallasii*, but I find a ready explanation in the fact that sand beaches are the rare exception rather than the rule along the Maine coast, so that the animal has simply accommodated itself to its environment.

Its home is a simple burrow formed by pushing aside the mud. The manner in which this is done was repeatedly observed both in its native haunts and in an aquarium.

If not already in that position, the animal turns until it rests upon its ventral surface. This brings the two large anterior setae in contact with the mud. The proboscis is now turned upward and backward, until it lies along the dorsal surface of the body, with its own ventral surface outermost, but protected somewhat by a rolling in of its edges. The proboscis remains inert in this position and *takes no part whatever in the burrowing*. This is in strong contrast to the active locomotor use of the proboscis described by Rietsch in a specimen of *Bonellia minor* (11). By a series of muscular contractions, which include both the longitudinal and circular muscles of the body walls and the special muscles which move the ventral setae, these latter are thrust forward until they project in front of the body almost horizontally.

At the same time the base of the proboscis is drawn backward and somewhat upward, so that the anterior end of the body becomes wedge or chisel shaped, the ventral surface being flattened and extending farthest forward, with the two setae projecting from its anterior edge. These setae are then turned downward and thrust into the mud as far as possible. Being curved, they furnish an excellent leverage, and the body is drawn toward them by a contraction of the longitudinal muscles.

This contraction passes slowly backward along the body until the posterior end is reached, which is moved forward

thereby half or three-quarters of an inch. The two anal rows of setae now serve to hold it in position, while the anterior end is again thrust forward and downward into the mud, and the ventral setae are fastened in a new position. This process is repeated until the animal finally disappears beneath the surface, leaving a circular opening equal in diameter to the body at its greatest lateral contraction. The whole process is extremely slow, and fully forty minutes are consumed in getting the posterior end of the body out of sight beneath the surface.

The burrow proceeds diagonally downward for ten to eighteen inches, then runs horizontally from six inches to two or three feet, and finally turns vertically upward again toward the surface.

When the animal reaches the surface the anterior end of the body is pushed out far enough to free the proboscis. This is then restored to its normal position and the body is withdrawn again into the burrow.

Spengel notes (15) that each burrow of the species (*E. Pallasii*) observed by him at Nordenay possessed two openings close together and each surrounded by a low wall. But those burrows were made in hard sand, while these are in soft mud, and consequently we should expect to find differences. These burrows at first have two openings, but the original entrance soon fills up through the caving in of its walls and the washing in of mud from the surface. The entire diagonal portion is often filled in this way and is never opened again, leaving this end of the burrow blind. These burrows also, when first formed, have low walls around the openings, caused by the pushing aside of the mud, but they quickly disappear.

The Echiurus assumes a position just below the surface, holding itself in place by the two rows of anal bristles (*cf.* Shipley). The mud then washes into the burrow and forms a plug one to two inches thick, with a small opening about the size of a lead-pencil at the center. Through this opening the proboscis is thrust out in search of food when the tide is in, and is then the only portion of the animal which is visible. It is capable of great extension, as was the proboscis of *B. viridis* described by Eisig, and often reaches a length of five or six inches. The

free end moves about in every direction and carefully searches the surface around the opening. Having found a particle of food, the edges are rolled in ventrally toward each other, if not already in that position, and form a more or less closed tube. The whole ventral surface (which is now the interior of the tube) being ciliated, there is generated a current which quickly carries the food toward the mouth. The proboscis often assumes a similar tubular shape when it is not elongated, as can be seen in Fig. 2, so that the curling inward of the edges is independent of the strong contraction of the circular muscles which produces the extension.

Often also it rolls itself into a tight coil, commencing at the tip and curling over ventrally as though it were grasping some object, but nothing save a few food particles is to be found in it, which are much too small to occasion any such effort.

The proboscis is very sensitive over its entire surface, but especially so on the ciliated ventral portion, and the slightest irritation there results in a quick withdrawal.

As would be inferred, such an appendage is of extreme importance to the animal, and yet it breaks off upon the slightest provocation. Whether such a separation is necessarily fatal or not, and whether the animal possesses the power of regenerating its proboscis, could not be definitely determined.

It hardly seems probable, however, that the animal could live for any length of time without it. But it was found that the proboscis itself was so highly innervated that it retained its vitality, and to a marked degree its sensitiveness also, for a long time after separation, a week or more if kept in fresh sea water. When the tide goes out, though there is always water left in the burrow, the proboscis is withdrawn and all indications point to the conclusion that the animal retreats to the lower part of its burrow.

Like other gephyreans, this species secretes a thick mucus, which lines the burrow walls and penetrates the mud for some distance, giving it greater firmness and solidity.

This mucus, as in so many other cases, oxidizes the iron in the mud, so that the walls of the burrow are a rusty brown color and stand out in sharp contrast to the surrounding black.

Movements and Locomotion. — In life, even when out of its burrow, the creature is constantly altering its outward form by energetic contractions of the skin muscles, as noted by Greef (6). Deep constrictions appear at various places, which move now forward, now backward, that portion of the body just in front of or behind the constriction increasing proportionately in size. The proboscis is also kept in constant motion, coiling up and uncoiling, rolling inward from the edges and unrolling or stretching out to a considerable distance and then being withdrawn. In its burrow the animal cannot turn around, but can move either forward or backward at will and with equal rapidity. This motion is accomplished by a series of wave-like contractions and relaxations in the circular and longitudinal muscles of the body wall, the one alternating with the other, and both together producing a fairly rapid gliding motion. The necessary rigidity is given to that portion of the body wall which for the time being serves as a fulcrum, by the pressure of the liquid in the body cavity, as first noted by Quatrefages (9). Andrews has clearly stated (1) the essential factors in the mechanism of *Sipunculus Gouldii* which bring about such "hydrostatic locomotion," and several authors have described similar motion in other geophyreans.

But no one, so far as known, has suggested any other mode of moving about. Indeed, one of the best recent text-books distinctly states that "the geophyrea are only capable of a very slow creeping motion" (Parker and Haswell, p. 461).

It seems never to have been suggested to any one, the present author included, that this same rhythmic contraction of the body walls would furnish an excellent means for swimming.

Hence it was quite a surprise, on visiting an aquarium after dark during the second summer, to find three or four specimens swimming about in it freely. The body was elongated to twice its ordinary length, while the proboscis was elongated even more in proportion and its edges were rolled downward and inward so as nearly to meet along the median line and form a long narrow tube which seemed to take an active part in the swimming.

The resultant motion was peculiar, being gyratory or cork-

screw-like, the anterior end always moving ahead, but it was perfectly free in any direction and quite rapid.

Besides assisting in locomotion the proboscis also seemed to serve as a steering organ, and its extreme sensitiveness rendered it very effective in avoiding obstacles.

The fact that this swimming took place only at night suggests that these animals are more or less nocturnal in their habits, and it may be that they can move about much more freely than has been hitherto supposed. At all events this is probably the mode of locomotion used at or near the breeding season, and it readily explains the large numbers of specimens which are thrown up on the beach after a storm at such seasons.

This species is well known to the clam-diggers along the coast and is sometimes used for bait in deep-sea fishing, but not often, and is never sought designedly for that purpose.

Determination of Species.—After a careful comparison of the descriptions given by Couthouy and Pourtales with that of *E. Pallasii* by its discoverer and subsequent zoölogists, and with the description which follows, there seems no possible doubt that those authors fell into the same error concerning our American species which trapped Quatrefages on the European form, *viz.*, they described an *Echiurus Pallasii* which had lost its proboscis as a new species. Accordingly *Holothuria chrysacanthophora* Couthouy, 1838, and *Echiurus chrysacanthophorus* Pourtales, 1851, must go to swell the long list of synonyms already appended to *Echiurus Pallasii* Guérin-Ménéville.

Echiurus Pallasii Guérin-Ménéville. — Synonyms: *Holothuria chrysacanthophora* Couthouy, 1838. *Echiurus chrysacanthophorus* Pourtales, 1851.

External Morphology.—Body like that of all known echiuroids, spindle-shaped, tapering slightly at either end; 10–30 cm.¹ long (including the proboscis, 3–6 cm. long) and 3–6 cm. in diameter at the center (Figs. 1 and 2).

¹ This figure is much larger than that usually given for *E. Pallasii*, but is the result of careful measurement and is good evidence in favor of Shipley's statement (13). "It seems probable that *E. forcipatus* of Reinhardt is identical with *E. Pallasii*, though bigger" (*cf.* p. 175).

Color uniform gray or grayish-yellow, shading into a deep orange on the interior of the proboscis. Entire surface of the body rough from being covered with small blunt papillae, which are unequal in size. The larger ones are more globular and are quite regularly arranged in transverse rows in which the individual papillae are so close together that they touch one another. There are twenty-two or twenty-three of these rows, and between them are scattered the smaller papillae, which are more conical in shape and seldom show any arrangement in rows.

Both kinds of papillae are more sharply defined and nearer together toward the ends of the body. There is also usually a bunching of the papillae around the anterior setae where they are larger than elsewhere on the body.

There is a pair of large, shining yellow, hooked setae, one on either side of the ventral mid-line, 16–20 mm. behind the base of the proboscis. These setae are about 20 mm. long and curve toward the posterior end of the body. They are retractile and can be almost wholly withdrawn into the body cavity. The posterior end of the body is surrounded by two rows of yellow setae, somewhat shorter than the anterior pair and perfectly straight. They also are retractile, and in most preserved specimens are withdrawn into the body cavity. In the animal shown in Fig. 4, however, they were extended to their full length. The rows are quite near together (3–5 mm.) and not more than 4 mm. from the anus, which is central and terminal. These posterior setae incline backward and assist the animal in moving about. The anterior row is made up of eight or nine setae, the posterior one of seven or eight.¹

Reserve setae are present for both rows and for the ventral pair. The setae alternate in the two rows, but neither row is entire, a space being left on the ventral mid-line.

In the posterior row this space corresponds to the omission of one seta, in the anterior row to the omission of three.

There is a papilla around the base of each seta, much larger than those on the body. The spaces between these basal papil-

¹ The fact that these different numbers may be found in individuals otherwise exactly alike is still further evidence that Reinhardt's species, *forcipatus*, is not well grounded.

lae and the intervals on the ventral surface, up to within 1 mm. of the mid-line, are filled in with ordinary large papillae.

Proboscis large, 3-6 cm. long and 1-3 cm. wide. It is capable of being extended to 12 cm. with a corresponding diminution of its width. It is cylindrical at the base, but quickly opens into a half tube which is broadened at the tip into a shovel form (Fig. 1). The exterior is a brighter yellow than the body and perfectly smooth. The interior is rich orange and completely ciliated, but in most specimens examined it lacks the longitudinal brown stripes noted by Greef in *E. Pallasii* (6). In several specimens, however, they showed up faintly against the orange background.

The skin also on the interior of the proboscis is thrown up into longitudinal ridges, the intervening furrows between which are darker in color than the ridges themselves (Fig. 3). The mouth opens through the center of the cylindrical base. A well-defined ridge runs outward from the mouth along the dorsal mid-line toward the tip of the proboscis. This ridge is somewhat brighter in color than the rest of the interior, but is completely concealed unless the proboscis is opened. The tip of the proboscis sometimes has a well-marked chocolate-brown edge.

The ventral nerve cord and blood vessel show plainly through the skin (*cf.* Figs. 1 and 2), and when the body is extended the dark-colored intestine can be seen at points where it touches the inner surface of the body walls. The sexes are alike externally, with no appreciable difference in size (*cf.* Figs. 1 and 2).

But when sexually ripe, Greef says that the golden eggs or the white semen in the nephridia show through the body wall enough to distinguish the males from the females (6).

Internal Morphology like that of all echiuroids. The alimentary canal is several times the length of the body and is looped irregularly. It is suspended from the body walls by thin muscular strands instead of a continuous mesentery, and upon the slightest perforation of the body walls it is extruded through the orifice by a violent contraction of the muscles.

This alimentary canal may be divided into three regions or parts, called respectively the fore, mid, and hind gut.

The foregut is very short, and contains a pharynx and oesophagus which are often bright orange in color like the inside of the proboscis. The remainder of the foregut is usually larger in diameter and has been called the crop. The midgut constitutes the bulk of the alimentary canal and is distinguished by a groove lined with vibratile cilia which runs along its dorsal side. Opening into this groove at either end is a collateral intestine, much smaller in diameter than the midgut and seemingly analogous to that in echinoderms.

The hindgut is somewhat larger than the midgut and forms near the anus a cloaca into which open two anal vesicles, one on either side. These are quite long, simple sacs, light brown in color, which vary greatly in length in different individuals (40-70 mm.). They open into the body cavity by ciliated funnels, which are most numerous near the base of the sacs, and one of which is terminal.

Both males and females have two pairs of nephridia, which open on the ventral surface on either side of and close to the nerve cord. The mouths of these nephridia are raised into large papillae on the external surface of the body, and can be plainly seen, the anterior pair just behind the ventral setae, and the second pair 5 or 6 mm. farther back.

At the base of each nephridium on the inner side is a ciliated funnel opening into the body cavity, through which the sexual products enter the nephridium when sufficiently ripe.

They are then discharged through the external papillae into the water. When free from eggs the nephridia are 15-20 mm. long and spindle-shaped, with a diameter of 3-5 mm. at the center. Probably when filled with ripe eggs or sperm they increase proportionately in size.

The *Sexual Products* are doubtless formed, as stated by Greef, from small cells near the posterior end of the ventral nerve cord, which are covered with peritoneum. But reproduction certainly does not take place in this locality (Casco Bay) in July and August, as well as in midwinter, *viz.*, it does not occur twice a year. All my specimens were secured in June to August, and not one of them contained ripe sperms or eggs.

But two specimens were obtained September 4 and placed in an aquarium. One of these, which proved to be a female, was injured in the digging, and while being put into the aquarium some nearly ripe eggs escaped through the rent in her side. These furnished the necessary stimulus for the uninjured male and he soon sent out ripe sperms in large quantity from the nephridia.

This would indicate a breeding season for that locality of September to November.

In the female just mentioned the nephridia were perhaps one-eighth full of eggs; all the rest of the eggs were free in the body cavity. This was the only specimen in which any eggs were found in the nephridia, but they may sometimes be found in the body cavity in June, and probably require a long time for development. When ripe (*i.e.*, those from the receptacles) the eggs are spherical, about 0.3 mm. in diameter and nearly opaque, but until fertilization the large germ nucleus can be plainly seen through the yolk granules.

The spermatozoa have a peculiar bullet-shaped head, a short cylindrical middle piece, and a long, very delicate tail. Their vibratory movements are rapid and very strong, and they retain the power of motion for a long time after being discharged into the water.

Just enough description has been here given to fix the species definitely, but considerable work has already been done on the morphology and histology of the body organs and on the origin of the sexual products, and it is expected that the near future will afford an opportunity for a careful study of the complete life history of this interesting species.

Through the courtesy of Dr. W. R. Coe, of the Sheffield Biological Laboratory at Yale University, I have received specimens of an *Echiurus* secured by him in Alaska, while on the Harriman Alaskan Expedition during the summer of 1899.

This species was found abundantly at many different localities along the Alaskan coast south of the Peninsula and on adjacent islands, nearly always in rich black mud.

It is of considerable interest to note that it proves to be the same species here described, *viz.*, *Echiurus Pallasii*, and that

its habits, so far as observed, correspond exactly with those just given. Its burrow is horseshoe-shaped, the two ends opening at the surface, and around each is a little mound formed by the pushing aside of the mud. The iron ingredients of the mud in the walls of the burrow are also discolored by the mucus secreted by the animal and show as a rusty brown.

In size the Alaskan specimens surpass those from Casco Bay, and the same shovelful of mud often reveals giants and pigmies of the species side by side. But this is simply in accordance with the general results of the expedition, for gigantic specimens of nearly every native species were found.

The number of setae in the anal rings of four specimens selected at random were counted. In three of these there were eight setae in the anterior ring and seven in the posterior, but in the fourth specimen the numbers were nine and eight respectively.

The fact that specimens from two such widely separated localities agree perfectly in carrying the maximum of size beyond 30 cm. and also in the variation of the number of setae in the anal rings, is a third argument, and quite a strong one, against the validity of the species *forcipatus*.

There seems to be no discernible connection between the number of the setae in the anal rings and the size of the animal; a small specimen is just as likely to possess the larger number.

On the contrary, there is something of a connection between the size of the individual and the temperature of its environment; in general, the colder the water the larger the average of the species. Such a fact strongly corroborates the statement made by Shipley (14) that "this genus is a denizen of the colder seas," and indicates that an Arctic environment is most congenial to its development.

These two new localities also go far toward rendering this species cosmopolitan. It has already been reported from the North Sea, where it was originally discovered, the English Channel, and the coasts of Norway, Sweden, Denmark, Holland, and Belgium. To these can now be added the American North Atlantic and North Pacific, and it may be expected as

one of the results of further investigation in the Asiatic North Pacific.

Thalassema erythrogrammon Max Müller.—I obtained a specimen of this *Thalassema* through the kindness of Dr. E. A. Andrews of Johns Hopkins University.

It was taken at Green Turtle Cay, off Great Abaco Island, the Bahamas, in the summer of 1886, and when alive was of a flesh color with reddish longitudinal stripes, the proboscis lighter in color, the papillae whitish. The body was raised into longitudinal ridges between the muscle bundles whose prominence varied with the degree of contraction. The specimen was hardened in Perenyi's fluid, and yet the muscle bands show a decided pink-brown color at the present time and stand out very distinctly, as can be seen in the photograph (Fig. 5).

It was excellently preserved in a normal condition and measures 16 cm. in length (including the proboscis, 3 cm. long) and 2.4 cm. in greatest diameter. Body spindle-shaped, with bluntly rounded ends, and after preservation *not* perceptibly furrowed by the longitudinal muscle bands. Papillae in dense plaques at the posterior end of the body; no smooth area in the immediate vicinity of the anus. Longitudinal muscles in sixteen bands about 1.5 mm. wide, with interspaces 4.5–7 mm. wide at the center of the body, except the two bands on either side of the ventral mid-line which are close together.

Proboscis so fleshy as to be nearly a solid cylinder, 1 cm. in diameter at the base, thus bringing the mouth to the extreme ventral surface; less fleshy and not broadened toward the tip.

The two setae were so far withdrawn into the thick skin as to be wholly invisible from the external surface, but could be all the more plainly seen on the interior.

The specimen proved to be a ripe male, and the three pairs of nephridia were enormously swollen and packed with sperm.

They increased in size from in front backward, the respective lengths being 3.2, 4.5, and 8.2 cm. The two posterior pairs were constricted at intervals and looked much like a string of sausages; the anterior pair opened 3 mm. in front of the ventral setae, and all three pairs were furnished with spirally coiled internal openings.

Anal glands 9 cm. long, simple, very thin walled, and *without* visible funnels. Intestine filled with the powdered shells of small lamellibranchs.

The chief interest in this specimen centers in the new locality. It has been reported hitherto only from the Red Sea, the Isle of Bourbon, the Malay Peninsula, and New Guinea, about as far distant as possible from the Bahamas. But it evidently belongs to the West Indian fauna and adds one more to the Atlantic species of this genus. Again, this is one of the species in which the number of muscle bands has been given as invariable and fourteen in number. The occurrence of sixteen bands in the present specimen shows that, like most of the other species, the number varies within narrow limits. The position of the anterior nephridia in front of the anal setae, as in *T. caudex* Lampert, is also worthy of note.

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WESTFIELD, MASS.,
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LITERATURE CITED.

1. ANDREWS, E. A. Anatomy of *Sipunculus Gouldii* Pourtales. *Stud. Biol. Lab. Johns Hopkins Univ.* Vol. iv.
2. CONN, H. W. Life History of *Thalassema mellita*. *Stud. Biol. Lab. Johns Hopkins Univ.* Vol. iii. 1884-87.
3. COUTHOUY, J. P. Description of New Species of Mollusks and Shells. *Bos. Journ. Nat. Hist.* Vol. ii. 1838.
4. FORBES AND GOODSIR. Natural History of *Thalassema* and *Echiurus*. *Edinburg New Phil. Journ.* Vol. xxx. 1841.
5. GOULD, A. A. Report on the Invertebrata of Massachusetts. Boston, 1841.
6. GREEF, R. Die Echiuren (*Gephyrea armata*). *Acta Ac. German.* Vol. xli, pt. ii. 1879.
7. GREEF, R. Ueber Echiuren und Echinodermen. *Archiv für Naturgesch.* 46 Jahrg. 1880.
8. POURTALES, L. Gephyreans of the Atlantic Coast of North America. *Amer. Asso. Adv. Sci. for 1851.* Vol. v. 1852.
9. QUATREFAGES, M. DE. Mémoire sur l'Echiure de Gaertner. *Annal. des Sci. Nat.* Ser. 3, T. vii. 1847.

10. QUATREFAGES, M. DE. *Histoire Naturelle des Anneles*. T. ii, Géphyreens. Paris, 1865.
11. RIETSCH, M. Études sur les Géphyriens armés ou Échiuriens. Thesis, Geneva, 1886.
12. SELENKA, E. *Challenger Reports*. Vol. xiii. 1885.
13. SHIPLEY, A. E. Gephyrea and Phoronis. *Camb. Nat. Hist.* Vol. ii. London, 1896.
14. SHIPLEY, A. E. On a Collection of Echiurids from Loyalty Islands, New Britain, and China Straits, with an Attempt to Revise the Group and to Determine its Geographical Range. *Zoöl. Results*, etc. Camb. Univ. Press, pt. iii. 1899.
15. SPENGEL, J. W. Ueber die Organization des E. Pallasii. *Zoöl. Anz.* Bd. xl.
16. VERRILL, A. E. Recent Additions to the Marine Invertebrata of the Northeastern Coast of America. *Proc. U. S. Nat. Mus.* Vol. ii. 1879.



FIG. 1. — A male (right) and female (left) *E. Pallasii* in a normal state of contraction in sea-water. $\times \frac{1}{2}$. Photographed from life.



FIG. 2. — The same pair in the same position, but with the female contracted under irritation. $\times \frac{1}{2}$. Photographed from life.



FIG. 3. — Ventral view of proboscis and anterior body. Photographed from preserved Alaskan specimen. Life size.



FIG. 4. — Anal rows of setae. Photographed from preserved specimen. Life size.



FIG. 5. — *Thalassema erythrogrammon*. Photographed from preserved specimen. $\frac{2}{3}$ actual size.

SOME GENERAL FEATURES OF THE METAMORPHOSIS OF THE FLAG WEEVIL *MONONYCHUS VULPECULUS* FABR.

JAMES G. NEEDHAM.

I HAVE been for some time desirous of studying the development of some beetle which would represent metamorphosis in as complete a condition as is found within the order Coleoptera. Last summer I found an abundance of the flag weevil (*Mononychus vulpeculus* Fabr.) in all stages ; and this furnished me the opportunity for which I waited. The larvae of this beetle are little fat grubs, which eat the seeds of the blue flag (*Iris versicolor* Linn.). They are sheltered from first to last within the flag capsule and are very degenerate. They lack eyes, antennae, and legs, as well as wings. They represent a sort of ecological specialization, common among the higher insects, manifest in the adaptation of life to very special situations, and of life history to conditions of transient food supply.

I. *Life History.*

The life history of this long familiar species seems not to have been fully made known.¹ While gathering my material I was not seeking to determine the full life history, but now I find that my collections and notes reveal it pretty completely. Collected material gathered in at intervals of two or three days give data as follows: Eggs were first found June 8. The beetles had just begun to oviposit on the earliest of the flag flowers, first opened that day. Larvae were first found June 29, at which

¹ Dr. John Hamilton published fragmentary notes on its life history in 1894 (" *Mononychus vulpeculus* and its Parasites," *Entom. News*, vol. v, pp. 287, 288), describing oviposition and the form and feeding habits of the larva, and citing an instance of great destructiveness on the part of two parasites, *Pimpla inquisitor* Say and *P. pterelas* Say.

time but few eggs were hatched. Pupae were first found August 5, and newly transformed imagoes, August 8, two months after egg-laying began.

Examining my collection of several hundred larvae, after the manner of lepidopterists, I find among them three sizes of head, so distinct as to certainly indicate three larval stages. The first, which is of the size attained before hatching, measures in diameter .24-.26 mm., the second .40-.44 mm., and the third .81-.85 mm.

My notes and collection labels together indicate the following life history :

1. An egg stage, lasting about three weeks. The eggs lie at the bottom of punctures made through the wall of the flag ovary by the mother-beetle with her rostrum. The egg is pellucid white, broadly oblong-oval in outline, and measures .38 by .70 mm.

2. A first larval stage, lasting about five days. At the end of this stage an average larva measures 2.2 mm. in length by .4 mm. in greatest diameter.

3. A second larval stage, lasting perhaps ten days (certainly not over two weeks), at the end of which the larva measures 4.60 mm. in length by 1.02 mm. in greatest diameter. Thus far the larva remains slender and quite elongate. During these two stages it traverses the outer face of from three to five seeds, leaving a slowly widening, shallow, brown furrow across their surfaces.

4. A third larval stage, lasting a very little more than two weeks, and divided into two periods :

- (a) A period of feeding, and extraordinarily rapid growth, lasting hardly more than a week. The greater part of increase in size is attained during this short period. During it the larva is boring through the center of several seeds, feeding on their highly nutritive endosperm. At the end of it the larva measures 6.5 mm. by 2.5 mm.

- (b) A period of transformation to the pupa.

5. A pupal stage, lasting, apparently, not more than a week, spent within the larval burrow. The pupa is naked and smooth; except for a pair of recurved spines on the tip of the abdomen.

6. A period of adult life, lasting ten or more months. Of this time a month or more is spent (lasting until the bursting of the

flag capsules in autumn) quietly within the larval burrow; eight or more months are spent in hiding in winter quarters; activity only begins with the season of iris flowering, and lasts for about a month. Oviposition continues sparingly after the first week. Except at the beginning of the season, several developmental stages may be taken at the same time, and this fact renders dates of *first observation only* of value as indices of life history.

The more striking features of this life history are:

1. The small number of larval stages, for a representative of this order.¹
2. The exceedingly rapid growth during the first period of the third larval stage. That an animal which will live a year should attain the greater part of its growth within a week is indeed a striking phenomenon. To be sure, this growth is mainly increase of fat.
3. The long period of adult inactivity, extending through two stretches of warm weather.

The metamorphosis of this beetle is very complete. The segregation of the development life into growth period (period of partial anabolism — fat-making) and differentiation period is very marked. The transformation of the degenerate larva, lacking wings, legs, antennae, eyes, optic lobes, and salivary glands, into the adult with all these parts well developed, is very rapid. There are excellent reasons for believing that these things have been independently acquired in the order Coleoptera. Internal metamorphosis has as yet been studied only in such representatives of this order as, in the larval stages, have legs and antennae and eyes, and undergo a metamorphosis much less rapid and complete. Therefore, it should be important to learn whether this increasing periodicity in life history has produced the same changes here as in other orders, whether disappearance of larval appendages has resulted in the internal development of imaginal discs, whether rapid metamorphosis is accompanied by phagocytosis, etc.

The external signs of internal metamorphic processes begin to

¹ Dr. C. V. Riley found four larval stages in the clover leaf weevil. *Vide*, "The Clover Leaf Beetle *Phytonomus punctatus* Fabr.," *Rept. U. S. Dept. of Agric.* for 1881, pp. 171-179, Pl. X. The *Phytonomus* larva is less degenerate.

appear almost as soon as the larva is done feeding. There is a slight loosening of the cuticle, and contraction of the body away from it, especially on the dorsal side of the thoracic segments and of the head. The budding legs and wings, already visible



FIG. 1. — Full-grown larva of the flag weevil (*Mononychus vulpeculus* Fabr.). w, wing buds, and l, leg buds, as seen through the skin.

through the transparent skin (Fig. 1), begin to grow and extend downward. The fat at the interior end of the body begins to lose its mottled, slightly grayish appearance, and becomes homogeneous, translucent, slightly yellowish-white.¹

Then the larval skin is cast, and the pupa appears with all the adult appendages clearly recognizable.

After this the progress of internal changes may best be gauged by pigmentation; first, in the eyes, then in the tips of the hind wings, and lastly in the general integument. Some of the corresponding internal phenomena will be discussed under the following headings.

II. *The Hypodermis during Metamorphosis.*

The hypodermis of this weevil is not, so far as observed, destroyed during metamorphosis to be again rebuilt in any part. The cells, however, while maintaining fairly constant relations at their ends, externally with the covering cuticle and internally with the tenuous basement membrane, take on remarkable variety of form and show a fine capacity for shifting, massing, or scattering themselves, previous to the definitive formation of the adult chitin. This might have been anticipated, in view of the exquisite sculpture and ornamentation of the adult beetle. It will be of interest to consider first briefly the origin of the appendage buds ("imaginal discs").

Wings and legs appear at the beginning of the third larval stage, each as a slight thickening of the hypodermis, showing almost from the beginning a slightly concentric arrangement of the elongating cells. Fig. 2, B, shows the beginning of a middle

¹ Correspondingly it ceases to be stained black with osmic acid in fixation.

leg. A wing bud would differ only in having the inner surface of the hypodermis free from tracheae, etc.

The striking difference between the behavior of the cells in these buds, and the behavior of hypodermis cells elsewhere (observed, doubtless, by every one who studies sections of insect larvae), I have thought it worth while to emphasize in this figure. Fig. 2, *A*, shows the crumpling which precedes molting everywhere except in these buds. Fig. 2, *B*, is a bud, and shows instead the thickening of the hypodermis and the compression of its cells. Elsewhere the hypodermis stretches as it grows;

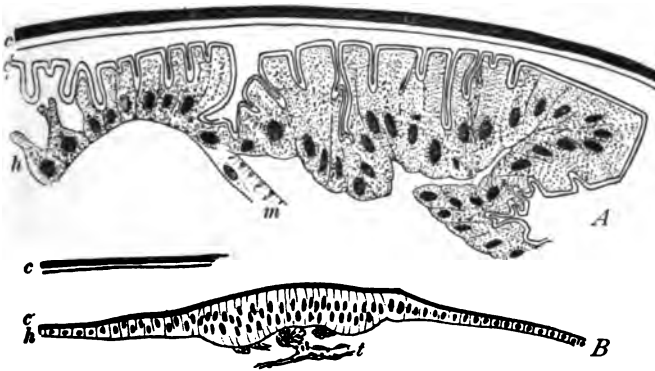


FIG. 2. — Two dispositions of hypodermis. *A*, vertical section of the frons at the end of the second larval stage, showing the crumpling of the hypodermis. *B*, vertical section of the bud of the middle leg at the time of molting at the end of the second larval stage, showing the thickening of the hypodermis, without crumpling. *c*, old chitine; *c'*, new chitine; *m*, developing muscle fiber; *t*, trachea.

the cells separate as they multiply; and that is true also of the hypodermis of the appendages, later, when the time has come for their extension. This we call retarded development, but the physiological explanation of it is still lacking.

There is no invagination of wing and leg buds in this beetle. Even the shallow hypodermal pockets formed about them in the more generalized Coleoptera are absent. The buds do not retreat from the surface. Fig. 1 shows their appearance as seen through the thin integument of the full-grown larva. Fig. 3 is a section of the wing at this time. The inner wall of the projecting shelf of hypodermis below the wing tip is all there is to represent the so-called "peripodal membrane." With

metamorphosis the wing begins to elongate from its apex and soon crowds downward past the shelf; and even before the casting of the last larval skin the general form of the adult elytron with the principal furrows upon its surface will have appeared.¹

The scales of this weevil are wholly developed during the pupal period. They vary in form from simple sensory hairs as

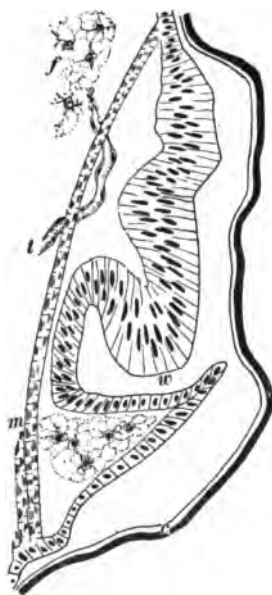


FIG. 3.—Vertical section of the fore wing in a grown larva. *w*, the wing apex; *c*, the loose chitine; *m*, muscle; *t*, trachea; *f*, fat.

in the antennal club, and slender, lanceolate, sensory scales in the tarsal brushes, to flat, longitudinally fluted, Lepidopter-like scales of yellow and black colors on the dorsal and more exposed surfaces, and delicate, short-plumose, white, or pale yellow scales on the less exposed surfaces. These, one and all, arise from ordinary, single hypodermis cells, after the manner of the development of the scales in the lepidopterous wings, as described by Mayer² *et al.*

First, in early pupal life the cells destined to produce the scales become much larger than their fellows and retreat a little from the surface, so that their nuclei appear at a lower level than the level of the other nuclei. Then each scale mother-cell loses its attachment to the basement membrane, becoming rounded off internally and sometimes acquiring a vacuole, and puts forth a process (the scale that is to be) between the adjacent surface cells (*cf.* Fig. 9). From this process the scale develops, the peculiarities of its own scale kind differentiating rather tardily.³

¹ There is no need to recount the wing development, since in all important features it is the same in this beetle as in a Coccinellid of which Professor Comstock and I have hitherto published an account (*Amer. Nat.*, vol. xxxiii, pp. 845–858, 1899).

² Mayer, A. G., "The Development of the Wing Scales and their Pigment in Butterflies and Moths," *Bull. Mus. Comp. Zool.* Vol. xxix, pp. 209–236, 7 plates, 1896. (Gives bibliography of earlier studies.)

³ By far the most interesting features of the hypodermis are found in the metamorphosis of the head and the development of the rostrum. These will

III. *The Development of the Legs.*

Aside from a few not very recent accounts of the development of the legs in Diptera, in which the leg buds are deeply invaginated in the larva, Gonin's account of them in the butterfly *Pieris* remains the only considerable one. And in *Pieris* there are larval legs, well developed and functional from the first. It



FIG. 4. — Longitudinal section of the middle leg in a grown larva. *f*, fat; *l*, leucocytes; *e*, embryonic cells. (Drawn from a preparation made in my laboratory by Mr. C. Betten.)

will be well, therefore, to notice here some points in the development of the legs of this weevil.

We have already called attention to Fig. 2, illustrating their origin. Fig. 4 is a longitudinal section of the leg of a grown larva, such as is shown in Fig. 1. Three principal divisions of the leg are already marked out by two deep constrictions. From the time of the beginning of the metamorphosis the growth of the legs is extremely rapid. Fig. 5, *A*, represents one of them as it appears after stripping off the larval skin just before pupation. (Fig. 8 shows wing and leg together, and in their relations to other parts.) The nine segments of the leg are constitute the subject of another paper, which is now being prepared by a student in my laboratory.

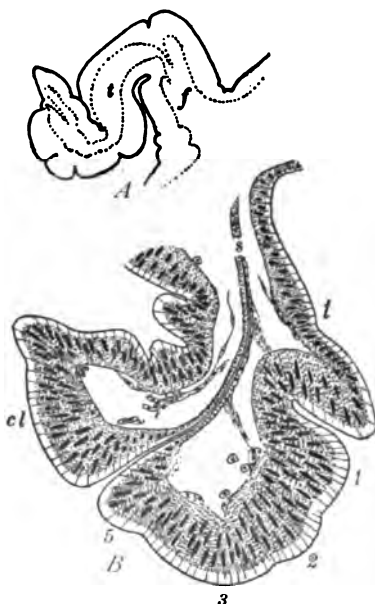


FIG. 5. — The leg of a larva, just before pupation. *A*, the entire leg in outline, and in part in optic section. *B*, a longitudinal section of the tarsus. *f*, femur; *t*, tibia; 1, 2, 3, 5, tarsal segments; *cl*, claw; *s*, developing tendon.

now clearly recognizable, all save one — the fourth segment of the tarsus, whose size is small in the adult, and whose suppression thus seems to extend back into the ontogeny of the species. Fig. 5, *B*, represents a longitudinal section through the tarsus at the same stage. This shows well the condition of the hypodermis at the time when all is ready for that great extension which accompanies pupation. Here is shown also the development of the strong tendon which protracts the claw, as a hypodermal invagination between segment five of the tarsus and the base of

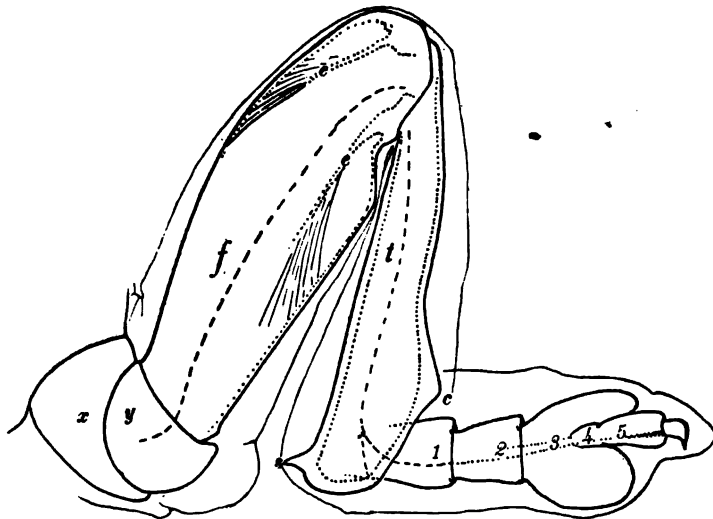


FIG. 6. — Leg of young pupa, within the pupal skin, in part in optic section. *x*, coxa; *y*, trochanter; *f*, femur; *t*, tibia; *c*, corbel; *s*, scrobe; *s*, *s*, developing tendons (flexor and extensor tibiae) and muscles; 1, 2, 3, 4, 5, segments of the tarsus.

the claw. At pupation this tendon is drawn out to great length, the hypodermis nuclei move apart, and spend themselves completely in chitine formation. Thus is the tubular ingrowth of soft hypoderm cells transformed into a solid cord of chitine. Muscle cells developing internally at the expense of the fat are from the first intimately associated with these hypoderm cells (*cf.* Fig. 9), and through them become attached later to the tendon. Just after pupation there is a striking similarity in appearance between these tendons growing from the surface into the leg cavity and the tracheae growing from the body cavity into it.

Fig. 6 shows the leg of a young pupa within its sheath. All the leg segments are rapidly assuming their definitive form. The fourth tarsal segment has reached its maximum development; it will be relatively smaller in the adult. The broad, flat, brush-bearing pads of the third tarsal segment are here big bag-like dilatations. Corbel and scrobe are very evident upon the tibia, and the femur and other segments are full of fat, rapidly being metamorphosed into muscle.

Fig. 7, *A*, represents the structure of the tarsus in an old pupa; externally it is practically that of the adult beetle. Fig. 7, *B*, is another section in the same series, passing through one of the lateral pads of the third tarsal segment. It shows a thinner hypodermis above, bearing scattered scales, and a thicker hypodermis below, bearing the dense tarsal brushes. Within are seen disintegrating fat cells, and other growing cells, angular and with large nuclei, which I take to be neuroblasts. Fig. 7, *C*, is

from a younger pupa. It shows well the manner of development of the tarsal brush. The mother-cells of its constituent scales settle below the general level of the hypodermis; owing to close crowding, their nuclei take on a cuneate form, and on the inner side of each a minute but distinct vacuole appears. At this age the hypoderm cells generally, as here, reach their basement membrane by long, peaked internal processes. Against this basement membrane here lie heaped embryonic cells, which later differentiate as the above-mentioned neuroblasts. Subse-

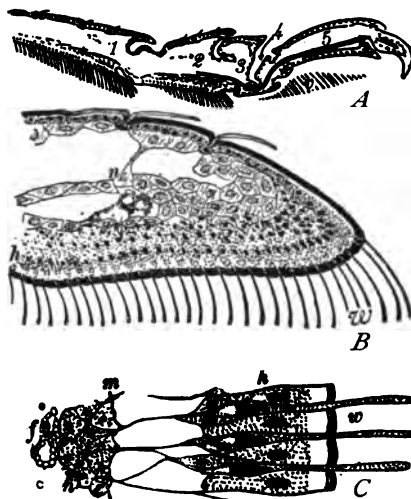


FIG. 7. — The tarsus in the pupal stage. *A*, longitudinal section of the tarsus in an old pupa. *B*, part of another section from the same series passing through one of the brushes of the 3d segment; *w*, the scales constituting the brush; *v*, the edge of one of the brushes belonging to the third segment; the tendon which retracts the claw is drawn in solid black; *n*, neuroblasts. *C*, a bit of a section through the tarsal brush in a young pupa, to show the origin of the scales; *w*, the scales; *h*, hypodermis; *m*, basement membrane; *n*, neuroblasts (undifferentiated); *f*, fat.

quent approximation of hypoderm cells and basement membrane (due, shall we say, to the drawing in of the peaked processes?), together with the loss of distinct cell boundaries in the hypodermis, renders the relation of parts much less clear in the later stages.

The tarsal claw and the tibial scrobe are developed alike from thick projections of hypodermis cells, forming at first a blunt point, which becomes sharp and takes on its characteristic tenacular curvature only when the chitine begins to harden. The corbel, however, being formed not at an angle of the leg, but upon an originally smooth surface, develops differently. There is a dense heaping of the hypoderm cells along what is to be the rim of the U-shaped corbel, among which the very large mother-cells of the fringing spines are early differentiated. Within the rim the cells are few, slender, and scattered. Outer (cuticle) and inner (basement membrane) surfaces are at first parallel; but the subsequent settling down of all the hypoderm cells upon their basement membrane leaves, where the few slender cells were within the rim, the proper concavity of the corbel.

IV. *Fat.*

The extraordinary growth taking place during the last larval stage is due almost wholly to the accumulation of fat. This occurring chiefly upon the dorsal side brings about the characteristic curvature of the larva. Hardly has growth been completed, however, before the reverse process sets in; the fat begins to be demolished and used in the construction of new parts. The external appearances accompanying the reduction of the fat have already been described. In sections the appearance is that of local disintegrations of the periphery of certain of the fat masses. Fig. 8 is a section through the middle of the thorax very near the beginning of metamorphosis. At the bases of the budding appendages and immediately above and below the alimentary canal, the fat is disintegrating. The appearance is that of the melting of frost. The fluid *residuum* flows forward into the head and laterally out into wings and legs, bearing along floating islands broken away from the fat masses.

During growth the fat is being stored in undifferentiated mesodermal cells which are free within the body cavity. These cells become greatly distended with the fat globules, which come to fill great interstices in the protoplasm toward the cell periphery. That each nucleus retains its vitality notwithstanding, is shown by its staining reactions, and by its retention of a central mass of protoplasm about itself, from which the peripheral strands that encircle the fat globules proceed. This is certainly not typical fatty degeneration; it seems to me much more properly considered to be partial anabolism, affected by these cells in their rapid elaboration of hydrocarbons during the transient period of abundant

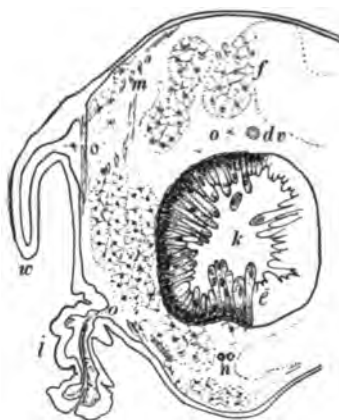


FIG. 8. — Partial cross-section of a larva, nearing pupation. *w*, fore wing; *l*, middle leg; *m*, muscles; *f*, fat; *dv*, dorsal vessel; *k*, alimentary canal; *e*, digestive epithelium, ready for dissolution; *n*, nerve cord; *o, o, o*, areas of first disintegration of the fat masses.

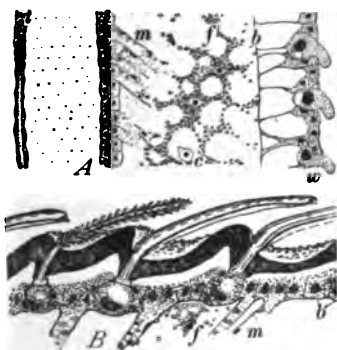


FIG. 9. — The development of scales and of muscle fibers. *A*, a bit of a longitudinal section of the femur of a young pupa; *t*, developing trachea; *s*, developing tendon (flexor tibiae); *m*, developing muscle fibers; *f*, disintegrating fat; *c*, a nucleus belonging to the fat mass isolating itself from the same; *b*, basement membrane; *w*, developing scales, in the midst of ordinary hypodermis. *B*, a bit of the body wall from a newly transformed imago, lettered as in *A*.

food supply. This view is corroborated by their later history. They do not (at least a majority of them do not) die with the dissolution of the fat. Nothing is plainer while one is watching the disintegration of the fat masses than that the nuclei contained therein show none of the usual signs of necrobiosis. Here and there will be seen a nucleus which, together with its enveloping coat of protoplasm, seems to be slipping itself free from its aforesaid accumulation of fat. Furthermore, these nuclei thus isolated can be seen associating themselves with the developing muscle rudiments, and, apparently,

themselves becoming the nuclei of new muscle fibers. The single fat globules which they often carry with them and sometimes retain, even after they have become associated with the muscle rudiments, enable one to follow them easily from their former situation into this new one.

There is no destruction of any larval tissue by phagocytes during metamorphosis, but *after the imago stage has been entered upon*, large numbers of phagocytes appear in the midst of the

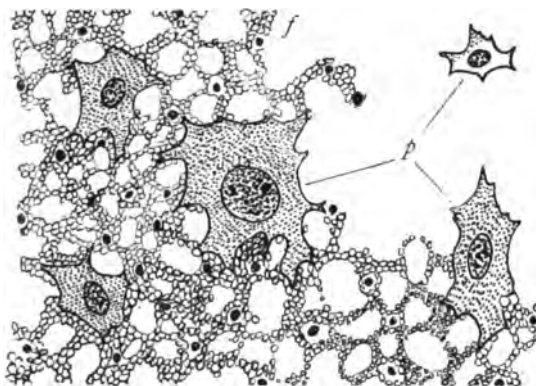


FIG. 10. — Phagocytes attacking the fat; the section is through the abdomen of a recently transformed imago. *p*, phagocytes; *f*, fat. (From a preparation made in my laboratory by Miss Elizabeth Andrews.)

fat along the sides of the abdomen. There are numerous embryonic or undifferentiated cells lying along the sides of the body in the larvae; and these, I believe, begin to penetrate the fat masses toward the end of the pupal stage. Fig. 10 shows the appear-

ance they present in a recently transformed weevil. Up to this time the fat filling the abdomen has not been greatly reduced, except in the anterior end; the change of form in the abdomen in passing from larva to imago is slight as compared with that of other parts. But it is clear that the internal metamorphosis is only well under way when the external is completed. This reserve store of fat is for the completion of the still weak organs of the imago, and for nutrition during the ten long months of inactivity remaining before the flags bloom and feeding begins again.

After calling attention to some of the interesting features of post-embryonic development, I would not close this little paper without mentioning the exceptional availability of this species for laboratory study. On a single trip to a favorable flag clump during the latter part of July in this latitude, one may gather in

a little while enough material for studying its entire metamorphosis. This material may be excellently fixed in boiling 70 per cent alcohol, and in all stages preceding the imago the chitine is so thin as to interfere but little with section cutting.

The following conclusions from the foregoing study are believed to be new:

1. In *Mononychus vulpeculus* Fabr. there are three larval stages.

2. The full-grown larva is very degenerate, having only the merest rudiments of antennae, eyes, optic lobes, and salivary glands.

3. The greater part of the increase in size takes place in about a week after entering the third larval stage; it is due mainly to fat accumulation.

4. This brief period of feeding and rapid accumulation of half-assimilated food material is correlated with an extremely long final assimilation period, lasting through months of imaginal life.

5. There is no real invagination of the buds of wings or legs.

6. Many nuclei of fat cells persist after the dissolution of the fat masses, free themselves from these masses, retaining about themselves an investment of protoplasm, associate themselves with developing muscle fibers, and, probably, themselves become the nuclei of new muscle fibers.

7. Phagocytosis, which was observed only in the fat masses along the sides of the abdomen, occurs only after external metamorphosis is complete.

NOTES ON THE PHYSIOLOGY OF REGENERATION OF PARTS IN PLANARIA MACULATA.¹

C. C. LEMON.

I. *Modes of Regeneration.*

In *Planaria maculata* there are two methods of inducing regeneration. First, isolated parts of sufficient size taken from any part of the body except the region in front of the eyes will regenerate; and second, partly isolated areas may regenerate, producing compound planarians.

1. *Isolated Parts.*—Randolph,² 1897, states that when a worm was cut into eight pieces by cross cuts, seven of them lived, and six of them regenerated all lost parts. The seventh failed to regenerate eyes.

Morgan³ has shown that there is a limit of size below which regeneration of lost parts will not take place. He also thinks that while the area in front of the eyes, which does not regenerate, is near this lower limit of size, there is another cause, probably that of greater specialization, why it will not regenerate lost organs.

My own observations on regeneration of isolated parts, though limited, for the most part support those of Morgan, as will be seen from the following record of experiments. A worm 10 mm. long and 2 mm. to 3 mm. in width was cut into

¹ The work herein recorded was done in the Laboratory of Experimental Morphology of Michigan University, under the direction of Dr. F. R. Lillie, to whom the writer wishes to express his sincere thanks for assistance and encouragement.

² "Observations and Experiments on Regeneration in Planarians," Separat-Abdruck aus dem *Archiv für Entwicklungsmechanik der Organismen*. Bd. v, p. 355. 1897.

³ "Experimental Studies of the Regeneration of *Planaria maculata*," Separat-Abdruck aus dem *Archiv für Entwicklungsmechanik der Organismen*. Bd. vii, pp. 365-372. 1898.

eight pieces as nearly equal in size as possible. All pieces regenerated lost parts and became fully developed worms in about ten days at ordinary room temperature. Another worm 5 mm. long and 1 mm. wide was cut into eight pieces. The operation was, however, so delicate that there was not much certainty in obtaining uniform size of the pieces. The larger pieces regenerated the lost organs, while the smaller ones did not. Just what the limit is, was hard to ascertain, as the relation of the piece to the whole could not be accurately determined, on account of its constantly varying shape. Parts which, by the most careful measurements, were shown to be about one-twelfth of the size of the original animal, regenerated and became fully formed planarians, while those of smaller size did not. Experiments on sixteen worms resulted in the same way. The area in front of the eyes did not regenerate in a single case.

2. *Production of Compound Planarians.*—This may be brought about in two ways: (1) Parts separated by cuts made along or near the middle line will generally complete themselves by regeneration without much growth. (2) Even extremely minute strips partly isolated may grow out like buds, and when of sufficient size, develop the characteristic organs of the species.

There is, of course, no line of demarcation between these two ways, which are united by a series of intermediates.

a. *By Regeneration.*—When a worm was split through the middle line of the anterior part of the body, sometimes the partly isolated left half regenerated a new right half and the partly isolated right half a new left half, thus producing a worm with two complete heads (Fig. 1). A similar operation may be performed on the posterior part of the body, resulting in two tails (Fig. 2). The time required for the regeneration of two heads is fifteen to twenty days, varying somewhat according to temperature. The regeneration of double tails occurred in five to ten days.

On Dec. 24, 1898, a large planarian was operated on by splitting the tail, as indicated in Fig. 13, except that the cut did not extend through the pharynx but only to the region

just posterior to it. On Jan. 3, 1899, two fully formed tails had developed. On January 5, the animal divided by fission about 3 mm. in front of the point of union of the two tails. The part possessing the two tails regenerated a new head, producing the animal seen in Fig. 3.

On January 11, the anterior part of the worm was again split posteriorly, this time dividing the pharynx. Five days later, on January 16, the posterior end of the worm again divided off, and subsequently regenerated a new head, as seen in Fig. 4.

The third and most anterior part of the original worm was split posteriorly, but the double tails

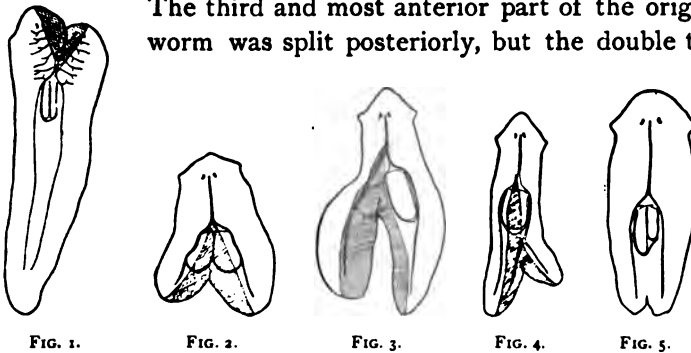


FIG. 1.

FIG. 2.

FIG. 3.

FIG. 4.

FIG. 5.

FIG. 1. — A double-headed planarian caused by regeneration after the original head had been split.

FIG. 2. — A planarian with two tails resulting from splitting and regeneration.

FIG. 3. — A planarian which separated from that of Fig. 5 by fission after its tail had been split.

FIG. 4. — This worm also separated from that of Fig. 5 by fission eleven days after Fig. 3.

FIG. 5. — The head end of the worm from which those in Figs. 3 and 4 separated.

could not be produced again, although the operation was performed three times. The only result that could be obtained was a worm with a slightly bifid tail and double pharynx (Fig. 5).

An interesting feature of the experiment with this worm is the way in which the alimentary canal developed in the regenerated parts. The left tail of Fig. 3 is supplied with nutriment by means of a sub-branch which comes off from the anterior branch of the canal, while the original left branch of the canal, which was severed in the operation, has disappeared. On the other hand, the right tail of Fig. 4 receives its nutriment by a sub-branch from the right posterior branch of the canal, which still persists, or is possibly a new formation. This and other problems concerning the anatomy of compound planarians are of interest and should be worked out.

b. *Budding*. — Small strips of tissue from the margin of the body or edge of a cut resemble buds in their capacity for growth and differentiation. These false buds regenerate more rapidly than larger portions of the body. To induce the growth of buds an incision is made, partly severing a very narrow strip (.5 mm.) of tissue, as shown by the lines in Figs. 6 and 10; Fig. 6 *a* indicates the method by which the worms in Figs. 8 and 9 were produced, and Fig. 10 *a* indicates how Figs. 11 and 12 originated.

In Fig. 7 the cut was made as indicated by the dotted line *a*. The bud, which was 4 mm. long, regenerated a new head, with brain, eyes, and cephalic lobes, in fourteen days. This head was developed from tissue in the posterior third of the body. Dalyell,¹ referred to by Randolph, thought that heads could be developed from tissue of the anterior part of the worm only. This idea is wholly disproved by Figs. 7, 11, and 12. At *b*, Fig. 7, is seen a bud one day after being cut.

i. *Growth*. — The bud, not having sufficient muscular strength to right itself against the larger part of the worm, heals without uniting with it, as is the case so often with animals split in the middle line. Growth begins very soon after the operation, being quite perceptible at the end of two days. It occurs in two ways: first, by regenerating new tissue on the cut edge of the bud; and second, by the increase of length, breadth, and thickness of the old tissue.

ii. *Differentiation of New Organs*. — Along with the increase in size the body becomes rounded off on the dorsal surface, and the head becomes broader and thicker in the region of the brain area when the cephalic lobes appear. Finally the eyes and pharynx, where a pharynx is developed, appear almost simultaneously.

In Fig. 8 the bud was formed by partly isolating a narrow strip of tissue from the side of the anterior part of the animal, as indicated by the shaded part *a*. About the time the cephalic lobes appeared, which was twelve days after the operation, the bud began to assert its independence, and was dragged about by the stronger worm with its head extending in a pos-

¹ "Observations and Experiments on Regeneration in Planarians," p. 370.

terior direction. As a result of the tension caused by the pulling, growth took place in the region *a* (Fig. 8), making the position of the posteriorly directed head a permanent and natural one.

In the case of Fig. 9 the bud was produced in the same way, but from day to day as the tension increased a slight cut was made at *a*, and as a result we do not have the head end of the bud directed posteriorly to the main axis of the worm, but nearly at right angles to it. The cutting prevented growth, and hence, when the animal comes to rest, or when relaxed in

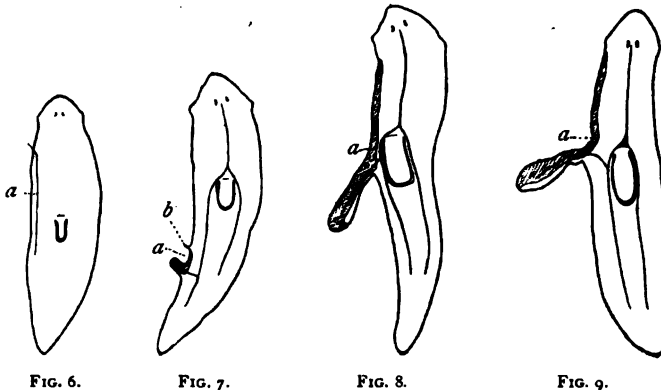


FIG. 6.

FIG. 7.

FIG. 8.

FIG. 9.

FIG. 6. — At *a* is seen the kind of cut which was made to produce buds.

FIG. 7. — A head regenerated from a bud, and a bud, *b*.

FIG. 8. — Pseudoheteromorphosis.

FIG. 9. — Frequent cutting at *a* prevented pseudoheteromorphosis in the worm of this figure.

killing, the bud, instead of remaining in a posteriorly directed position as when in motion, takes a position more nearly the same as that which it originally occupied.

While the bud was developing, the cut edge of the larger part regenerated enough new tissue to replace that which went to produce the bud. Thus we have a well-formed double-headed planarian in the case of Fig. 8. In Fig. 9 the bud failed to develop a left eye. This may be due to the frequently cutting at *a*; otherwise our present knowledge of the case makes it impossible to decide what the cause may be.

iii. *Final Fate of Parts.* — One point was quite noticeable in all the experiments with buds. When the animal had be-

come well formed there was a strong tendency to divide and in this way get rid of the abnormal condition. Ignorance of this fact cost the writer two of his best examples of regeneration; and it was only by diligently watching their development and killing the material at the proper time that the examples for this part of the paper could be obtained.

II. *Heteromorphosis.*

1. *Historical.* — Randolph¹ mentions four cases found by Dalyell in 1811 which are worthy of mention here. The first was a planarian with a bifid tail, between the two branches of which was an erect structure supporting a head. Second, a planarian, upon the side of which incisions had been made, developed a head pointing downward in the direction of the tail. The third and fourth cases consist of two monstrosities, the description of which is quite similar to that of Fig. 11. These were two worms, each of which had another attached to it and lying at right angles to its tail.

Van Duyne² gives three figures which he claims prove the possibility of heteromorphosis in the planarian. One, his Fig. 3, represents a worm with two heads on the anterior part of the body, one of which points posteriorly. The second one, his Fig. 4, shows a worm whose body has been split through the tail almost to the head. Between the two tails thus produced two heads have appeared, which, when the tails are widely separated as represented in the figure, point in a posterior direction. And lastly, his Fig. 5 represents a tail pointing in an anterior direction.

Morgan³ gives one example of apparent heteromorphosis, his Fig. 36. It shows a worm with two heads, which point

¹ "Observations and Experiments on Regeneration in Planarians," Separat-Abdruck aus dem *Archiv für Entwicklungsmechanik der Organismen*. Bd. v, p. 367.

² "Ueber Heteromorphosis bei Planarien," Separat-Abdruck aus dem *Archiv für die ges. Physiologie*. Bd. lxiv, Taf. x.

³ "Experimental Studies of the Regeneration of *Planaria maculata*," Separat-Abdruck aus dem *Archiv für Entwicklungsmechanik der Organismen*. Bd. vii, p. 381.

in opposite directions when in a relaxed condition, but when expanded form an angle of about 100° .

Morgan¹ has confirmed Spallanzani's discovery of earthworms regenerating a tail in place of a head. Sections of these worms show a ventral cord extending to the new part, that no brain is present, and that the nephrostomes in the new part are turned backward towards the old part.

Loeb,² in his investigations to determine the cause of animal forms, produced monstrosities with hydroids in which the oral end was regenerated on the aboral end. Loeb proposed the term "heteromorphosis" for such monstrosities. Heteromorphosis not only includes the regeneration of a head in the place of a tail, but of any organ in any place where in nature one of unequal value would occur, as arms from the hips and legs from the shoulders, etc. Loeb defines heteromorphosis as "the replacement of one organ by another physiologically and morphologically different."

2. *Analysis.*—The term "heteromorphosis" thus includes the entire reversal of axial relations as well as the development of any single organ in place of another. It will be useful to distinguish these as polar heteromorphosis and heteromorphosis of single organs. Examples of the latter are found in various forms, as the regeneration of a tentacle-like organ in place of an eye in crabs, etc. Examples of polar heteromorphosis, on the other hand, with few exceptions, occur only among the Coelenterates.

Cerfontaine,³ in his "Observations physiologiques sur l'*Astroides calycularis*," records the regeneration of a crown of tentacles on the base of a severed part of a polyp.

Loeb has found axial heteromorphosis to be quite common among the coelenterates and has been able to produce it in

¹ "A Confirmation of Spallanzani's Discovery of an Earthworm Regenerating a Tail in place of a Head," Abdruck aus dem *Anatomischen Anzeiger*. Bd. xv, pp. 407-410. 1899.

² *Untersuchungen zur physiologischen Morphologie der Thiere*. Bd. i, ii. Wurzburg, 1891.

³ "Notes préliminaires sur l'organisation et le développement de différentes formes d'Anthozoaires (deuxième communication)," *Bull. de l'Acad. Roy. des Sci., des Lettres et des Beaux-arts de Belgique*. No. 8, Notes v-viii. 1891.

at least the following forms: *Tubularia mesembryanthemum*, *Aglaophemia pluma*, *Plumularia pinnata*, *Eudendrium* (*rasimosum*?), *Sertularia* (*polyzonias*?).

Bickford and Driesch,¹ cited by Morgan, have also shown that in the tubularian hydroids two heads may develop on opposite ends of a piece cut from a stem, especially if the piece be short.

3. *Pseudoheteromorphosis*. — By cutting a narrow strip from any part of the body so as partly to isolate it, a posteriorly directed head may be developed by the reversal of the piece.

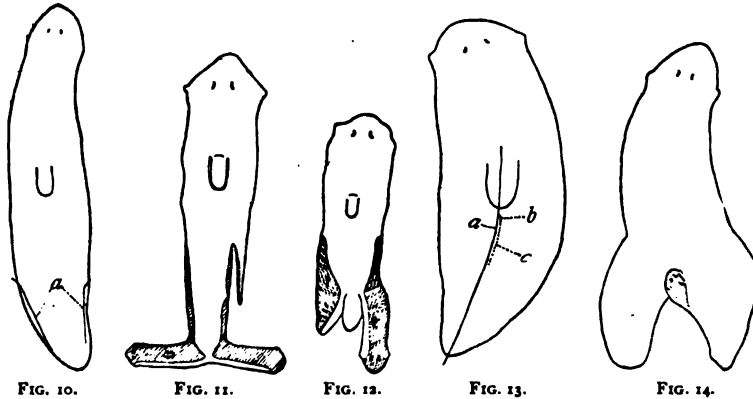


FIG. 10.

FIG. 11.

FIG. 12.

FIG. 13.

FIG. 14.

FIG. 10. — The lines *a* show the nature of the cuts which produced heads at right angles to the body.

FIG. 11. — A worm with heads lying at right angles to the main body.

FIG. 12. — Pseudoheteromorphosis.

FIG. 13. — The lines *a*, *b*, and *c* represent the cuts made to induce regeneration of heads in the tail region.

FIG. 14. — A head regenerated on one of the tails.

If by tension and growth this position becomes permanent, forms are produced which, to the casual observer, appear to be marked examples of heteromorphosis. Figs. 8, 12, 15, and 16 possess all the outward appearances of true heteromorphosis, but by the aid of Figs. 6, 10, and 13 one can readily show that there is neither the reversal of axial relations nor the development of one organ for another. Hence we do not have true heteromorphosis, but simply the swinging around of a portion

¹ "Experimental Studies of the Regeneration of *Planaria maculata*," Separat.-Abdruck aus dem *Archiv für Entwicklungsmechanik der Organismen*. Bd. vii, p. 382.

of tissue as a whole, so as to give the anterior end a posterior direction, or pseudoheteromorphosis.

Perhaps the best example of pseudoheteromorphosis is found in Fig. 15, which was produced in the following manner. On Dec. 22, 1898, the worm was operated on by splitting its tail, as indicated by the line *a* in Fig. 13. Then a small anteriorly directed piece of tissue was partly isolated on the inner margin of the right tail *b*, which developed a head, as seen in Fig. 14, by Jan. 9, 1899.

Fig. 15 represents the same worm in an expanded condition during locomotion.



FIG. 15. — The worm of Fig. 14 in an expanded condition.

Fig. 16 was produced in the same way as Fig. 15, except that the strip, which was isolated from the inner edge of the right tail, was cut longer, as indicated by the dotted line *c* in Fig. 13.

Neither Van Duyne nor Morgan gives evidence of having produced anything other than pseudoheteromorphosis.

4. *Critique of Evidence.* — In Van Duyne's first example of heteromorphosis (Fig. 3 of the plate) he figures a worm with two heads, one of which arose from the wound caused by taking a piece from its side by two cuts, a transverse one back of the right half of the head, and a longitudinal one from the inner end of the first cut to the tip of the tail.

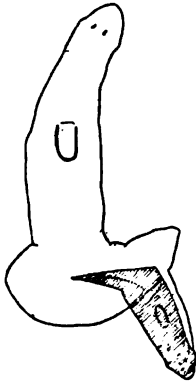


FIG. 16. — Pseudoheteromorphosis.

In order that this be an example of axial heteromorphosis it must have been regenerated from tissue which originally bore the same relation to the main axis of the worm as does the tail, *i.e.*, it must have been regenerated from tissue, the free end of which was originally posteriorly directed. The

drawing does not clearly show this, but rather indicates that this head may have been regenerated on the anterior end of the newly formed tissue on the side of the worm and was forced to turn backward by the shoulder-like projection of old tissue.

Likewise in Fig. 4 of Van Duyne's plate there is no evidence that the heads may not have arisen from anteriorly directed tissue, as did the head in my Fig. 15. Fig. 5 of Van Duyne's plate gives no more evidence of being a tail than of being a partially developed head.

Morgan, in Fig. 36 of his paper, gives an example of what he considers to be axial heteromorphosis. He says: "The entire history of this piece is known, and there can be no doubt that two heads developed on opposite ends of the same cross-piece." Further he adds: "The bending of the heads to one side is due, in all probability, to the knife cutting somewhat obliquely to the long axis at the time that the piece was removed." May it not be more probable that we have here a case of the development of a head from each of the anterior corners of the piece? It is certainly reasonable to suppose this in the light of the evidence given. To determine whether this be an example of axial heteromorphosis or not, two things are necessary, *viz.*: (1) That we know the end of the piece which was originally directed anteriorly by some means other than the direction of its motion; and (2) that we know that the same end, which was the anterior end when the piece was first cut, continues to be the anterior end of the newly developed worm. Several cases were noticed where the piece, either from not having been cut squarely across or from some other cause, at first moved in a direction diagonal to its antero-posterior axis, but afterwards, when the regenerated part developed normally, *i.e.*, in the line of the antero-posterior axis, it again moved in a straight line. If the new tissue developed a little to one side of the antero-posterior axis, as was sometimes the case, the piece continued to move in a diagonal direction, following the newly formed head. May not Morgan's Fig. 36 be an example involving conditions similar to these without involving axial heteromorphosis?

5. *Effect of Injury to One Part on a More or Less Different Part.*—In addition to the tendency to divide after the regeneration of new organs, referred to elsewhere, it sometimes happens that an operation on one part of the body produces an abnormality in some other part. Three interesting cases were

found where the eyes either divided, or became abnormally large and irregular in shape, and two where the pharynges developed abnormal proportions.

The first case was caused by an operation upon a planarian to produce a bud, as indicated in Fig. 6. Three times the bud divided off by fission, leaving the worm almost normal in appearance. After the third operation the eyes, which were crescentic in outline, began to deposit pigment in the concavity in irregular masses until the condition represented in Fig. 17 was produced, when the head separated from the body by fission.

The second case (Fig. 18) is that of a worm which had been operated on in a similar manner. The bud divided off and



FIG. 17.



FIG. 18.



FIG. 19.

FIG. 17.—A worm in which the eyes have become abnormally large after being operated on.

FIG. 18.—A worm in which two new eyes developed after an operation upon its sides.

FIG. 19.—The dark part of the right eye divided after operation on the side of the worm.

almost immediately the eyes divided, giving four eyes. The one on the extreme left side of the head has the concavity on the right side, suggesting the possibility of its functioning as a right eye. Two others are in almost the normal position, while the fourth lies between them and a little to the left of the middle line of the head.

The third case (Fig. 19) is that of a worm which had been operated on in the posterior part of the body on the left side, producing a bud. When the bud had become half grown it divided near its anterior end. The right eye of the worm then divided in such a way as to produce two, one lying just anterior to the other. The head of this animal also separated from its body by fission soon after the division of the eye.

The two cases of abnormally developed pharynges were

found in two worms which had been split near the middle line of the body. One was split through the head back to the pharynx but not including it. After several operations two heads developed, and it was noticed that the pharynx was gradually increasing in width. This continued until the two heads were fully formed, when it had reached a size nearly twice that of the normal.

The other case was a worm whose tail had been split to the base of the pharynx. After the operation the pharynx increased continually in width until two tails were fully formed.

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ANN ARBOR, MICH., July 28, 1899.

THE STRUCTURE OF THE EYE OF SCUTIGERA (CERMATIA) FORCEPS.

JOSEPHINE HEMENWAY.

GRENACHER ('80), in his article entitled "Ueber die Augen einiger Myriapoden," described the structure of the eye of *Scutigera* (*Cermatia araneoidea*). Briefly reviewed, his account is as follows:

Externally the eye of *Scutigera* has the appearance of a true facet eye, consisting of one hundred of these "facets." To each facet there corresponds an ommatidium. Each ommatidium consists of a central crystalline body, surrounded by three tiers of cells; the distal tier of pigment cells, the middle and proximal tiers of reticular cells secreting on their inner edges a narrow band, the rhabdom. The crystalline body is composed of very irregular segments. These segments may be either cells or cuticular structures. In the adult eye they cannot be regarded as cells, as nuclei are not found in them, although Grenacher admits that at some time in their existence they may have been cells, later becoming modified and losing their nuclei. The possibility of their being secretion products he does not admit, as he finds no cells to which their origin could be traced. There are six to eight or nine of these segments.

The reticular cells with their rhabdoms embrace the proximal two-thirds or three-fourths of the crystalline body, the posterior portion of the reticular cells reaching to the basal membrane. Of the three tiers of cells surrounding the crystalline body, the middle tier, or outer retinula, is made up of from nine to twelve cells; the proximal tier, or inner retinula, of three to four cells. Sections through the proximal layer show that at this level the rhabdom is made up of four parts. Toward the extreme proximal end of these proximal reticular cells only three with their rhabdoms are visible in cross-

sections, the fourth having been pushed out. The nuclei of the reticular cells lie in the distal portion. The pigmentation of the eye consists partly of the pigment granules in the reticular cells and partly in the separate pigment cells. Of the latter there are three distinct groups: (1) a circle of from eight to ten large flattened cells, the outer tier of my description around the outer part of the crystalline body; (2) long, spindle-shaped pigment cells situated between the ommatidia, extending to the inner cuticula; (3) a third group, the supplementary cells of my account, is found on the posterior part of the retinula, between the reticular cells.

Grenacher also mentions the pigmentation of the optic nerve and the "inner cuticula."

Adensamer ('93), in his studies on this eye (*Scutigera coleoptrata*), confirms and completes Grenacher's statements. He differs in certain points.

The cornea of each ommatidium Grenacher regarded as externally convex, although there were individual differences. These differences Adensamer regards as stages in the development of the cornea.

In the adult eye frequently there were found in the crystalline body large yellowish enclosures, which had the appearance of fat drops. These are not to be confused with the nuclei for which Grenacher looked. Of the segments he found from seven to nine.

But in an individual 5 cm. long Adensamer states that he found nuclei in the crystalline body; thus he feels justified in calling the segments "cells."

As to the nerve fibers he was more successful than Grenacher, in that he saw the connection of the fiber with the outer and inner row of reticular cells. This he proved by sections. Just under the basal membrane there is a nerve connected with a muscle, which is entirely distinct from the optic nerve. Adensamer believes that this was probably mistaken by Grenacher for the real optic nerve. The latter consists of a bundle formed of the separate nerve fibers meeting proximal to the basal membrane.

Speaking of the superficial resemblance of the eye of *Scutigera* to that of insects and crustaceans, and the actual differ-

ences between them, Adensamer suggests calling the eye of *Scutigera* a "pseudo-facet" eye. Rosenstadt ('96) discusses the question as to whether the eye of *Scutigera* can be regarded as a true facet eye, reviewing the arguments of Grenacher and Adensamer. He also suggests a way by which an eye, as that of *Scutigera*, could be developed from a true facet eye.

The following work was done in the Biological Laboratory of Bryn Mawr College, under the direction of Prof. T. H. Morgan, to whom I am greatly indebted for valuable suggestions and criticism.

The species studied was *Scutigera* (*Cermatia*) *forceps*.

For sectioning, the best results were obtained by hardening the fresh material in corrosive acetic for fifteen minutes, then running it up through the successive grades of alcohol.

The dense pigment obscured all details, therefore a depigmenting agent, as KOH, was used (*cf.* Parker, "The Eyes in Scorpions," '87). The preparations were stained with iron-haematoxylin.

As a maceration fluid, a modification¹ of Béla Haller's fluid was used. Material left in it for a year gave excellent results. The separate ommatidia fell apart, and by gently tapping the preparation the individual cells of each ommatidium could be isolated.

By this means I have been able to make out more definitely the structure of the different component cells than have the authors mentioned above, and in some respects have been able to add some points to their results.

The eye of *Scutigera forceps* is nearly triangular in shape. The corneal hypodermis is faceted, one ommatidium corresponding to each facet. Each eye is composed of about two hundred individual units or ommatidia.

Fig. 1 *A* shows² a single ommatidium, its proximal end bordering on the inner cuticula or basal membrane.

¹ Béla Haller's mixture modified: two parts glacial acetic acid; two parts water; one part glycerine.

² The figures are all camera drawings: Fig. 1 *A* and *B* were drawn with a No. 7 objective; Fig. 1 *C* and *D* were drawn with an oil immersion $\frac{1}{2}$; Fig. 2 *A*, *B*, *C*, *D*, *E*, *F* were drawn with an oil immersion $\frac{1}{2}$.

Surrounding each ommatidium are elongated pigment cells, extending the entire length of the ommatidium (Fig. 1 *B*). At both distal and proximal ends these pigment cells become expanded, the pigment granules collecting in the expanded portions. At the proximal end this gives the pigmented appearance of the basal membrane, spoken of by Grenacher.

There are sixteen to eighteen of these pigment cells belonging to an individual ommatidium. The nuclei are visible without reagents, but are more clearly shown by methyl green.

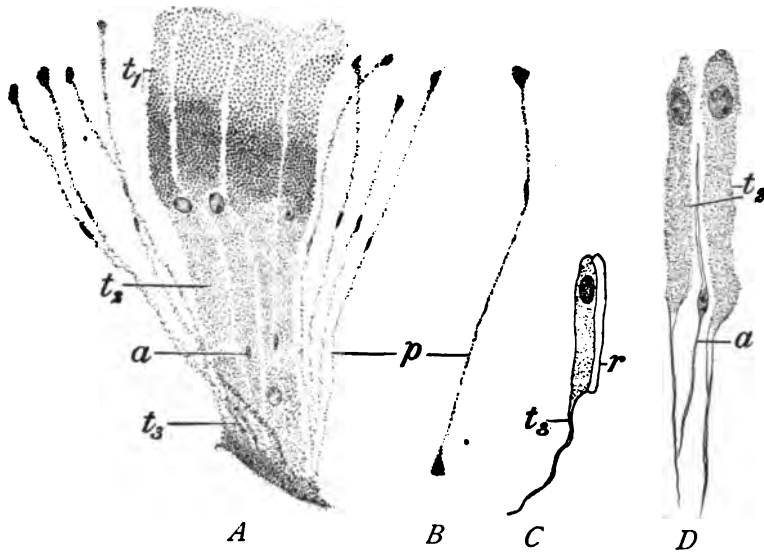


FIG. 1.

They are found at the same level as the nuclei of the middle tier of cells (Fig. 1 *A*). Each ommatidium consists of a clear, crystalline body, surrounded by three tiers of cells; the outer consisting of twelve (t_1), the middle of ten to twelve (t_2), and the inner tier of three to four cells, respectively (t_3).

The cells of the outer tier are large and flat and deeply pigmented at the proximal ends, the pigment granules being extremely large and round. The nuclei did not show in a macerated specimen, owing to the pigment.

The middle tier of cells are called by Grenacher the "outer reticular cells." They are longer and more narrow than the

cells of the outer tier, reddish in color, lacking the black pigment of the outer tier.

(Fig. 1) *D t₂* shows two of these cells with the nuclei at the extreme distal portion. At the proximal end each cell is prolonged into an extremely fine "tail," which runs down between the cells of the inner tier and is continued through the basal membrane as a nerve fiber.

The cells of the inner tier have their proximal ends bordering upon the basal membrane. From the proximal ends fine processes continue through the basal membrane to form the nerve fibers. The cells are much broader than those of the middle tier.

Cross-sections through the different levels show the crystalline body occupying the central axis of the ommatidium, surrounded by a clear zone or rhabdom forming the inner parts of the cells of the ommatidium (Fig. 2 *B*, *r*). The clear zone is formed of the structures called by Grenacher the rhabdoms—a secretion product of the reticular cells. In macerated specimens these rhabdoms were visible upon the inner surface of each cell of the two proximal tiers (Fig. 1 *C*) and could be made to separate from the cell by tapping. The "tail," or nerve, is on the opposite side of the cell from the secreted portion. The secretions from the inner tier are thicker than from the middle tier, and in cross-sections appear roughly triangular in shape (Fig. 2 *F*). There were no nerve fibers observed for the outer tier, and it differs in this respect from the two inner tiers. In certain cases, after tapping, the outer cells unfolded, as it were, and spread out into a band. The distal ends are rounded, while the proximal ends are drawn down into a point which extends between the distal ends of the middle tier. Thus the nuclei of the middle tier are found between these points of the outer tier (Fig. 1 *A*).

The series of cross-sections (Fig. 2 *A-F*) are taken at the levels of the different nuclei.

I. The first section beneath the cornea is shown in Fig. 2 *A*. It represents the extreme distal portion of the ommatidia. A few large nuclei are found at this level, situated between the individual ommatidia.

II. The next figure (Fig. 2 *B*) is taken through the nuclei of the outer tier of cells, two sections intervening between *A* and *B*. These nuclei differ in shape from the round ones of the middle tier.

III. The next section (Fig. 2 *C*) shows two sets of nuclei, the larger ones (*n*) belonging to the middle tier of cells, the smaller (*p*) being the nuclei from the surrounding pigment cells (Fig. 1 *A*, *p*).

IV. Fig. 2 *D* shows a section through the nuclei of the middle tier of cells. This drawing is from the same section as *C*, but drawn at a lower level.

V. The following figure (*E*) is the fourth section after *D*. The round nuclei are from the clear cells or supplementary cells.

VI. Situated at about the same level as the nuclei shown in Fig. 2 *E*, but in the next section (Fig. 2 *F*), are the nuclei belonging to the inner tier of cells (Fig. 1 *A*, *t*₃).

The nuclei shown in Fig. 2 *E* belong to a group of cells lying between the two proximal tiers of cells. These cells are very different from any of the others found. They are colorless and contain no granules. Fig. 1 *D* shows two cells from the middle tier, between them one of these supplementary cells, in its natural position. The supplementary cells are extremely delicate, sending distally a fine process between the cells of the middle tier, and proximally between those of the inner tier. The nucleus is small and easily distinguished from that of a cell of the inner tier. The nuclei are found at the level of the distal ends of the inner cells. There are four supplementary cells.

The crystalline body is surrounded by the tiers of cells described above. It is composed of segments — “Grenacher’s segments.” These segments are cone-shaped. At the level of the nuclei of the inner tier (Fig. 2 *F*) these are not seen in cross-section, the rhabdoms, only, belonging to these cells being visible. In maceration preparations they often separate from each other at the proximal ends, while found joined at the larger distal ends.

According to Adensamer there are nuclei in these bodies

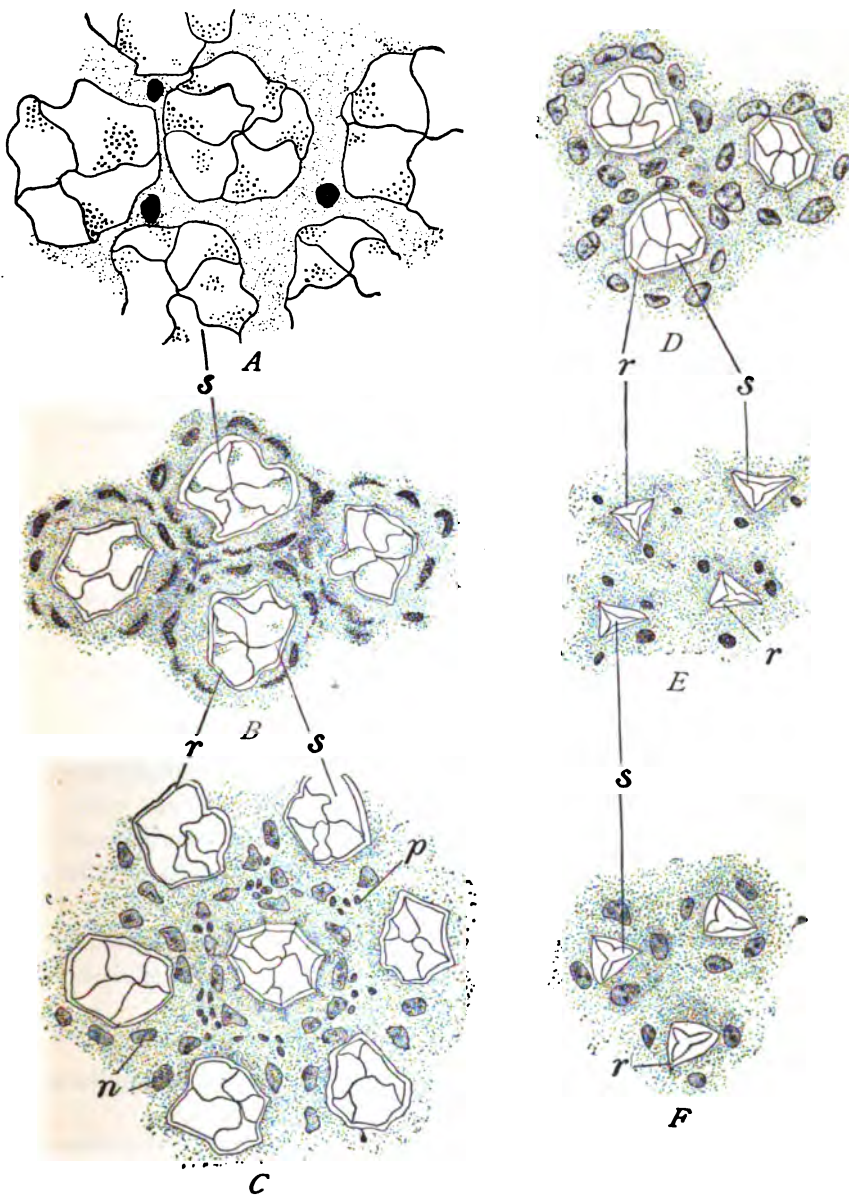


FIG. 2.

early in their existence, thus proving their cell nature. He also states that in an adult these can be vaguely seen. In no eye did I see an indication of nuclei in the segments of the crystalline body.

Cross-sections show the segments to be arranged in no regular manner. In a complete series of cross-sections one ommatidium was followed, and camera drawings at the high and low level were made of each section.

It was thus possible to trace each segment and find the number of segments and their relative position. In most ommatidia the number counted was ten to eleven, but in one ommatidium I was able to trace twelve. It must be understood that in a single section no more than six to eight appear. This can be seen by referring to Fig. 2.

SUMMARY.

The species described by Grenacher is *Scutigera* (*Cermatia* *araneoidea*); by Adensamer, *Scutigera* *coleoptrata*; by myself, *Cermatia* *forceps*.

The latter is the only American *Scutigera*.

The differences in the accounts are probably to be explained in part by the fact that the individuals studied were of different species.

(1) The number of ommatidia in each eye of *Scutigera* *forceps* is about two hundred. In *Cermatia* *araneoidea* (Grenacher) the number is given as one hundred.

(2) The crystalline body was found to be made up of ten to twelve segments, instead of six to nine. No nuclei were observed in these segments.

Each ommatidium is made up of the following cells :

(3) Elongated pigment cells surrounding each ommatidium, sixteen to eighteen in number (*p*, Fig. 1 *A*).

(4) An outer tier of pigment cells, embracing the distal portion of the crystalline cone, ten to twelve in number (*t*₁, Fig. 1 *A*).

(5) A middle tier of cells of ten to twelve (*t*₂, Fig. 1 *A*).

(6) An inner tier of cells situated at the proximal end of the

ommatidium, touching with their proximal ends the basal membrane. In the inner tier there are from three to four cells (*t*₃, Fig. 1 *A*).

The cells of (5) and (6) secrete, upon their inner surfaces, rhabdoms, and from the outer side send out a process constituting the nerve fibers.

These nerve fibers were mentioned by Adensamer, but his figures failed to show the direct connection between the fibers and the cells of the middle and inner tiers.

In macerated preparations I have been able to show this beyond doubt (Fig. 1 *C, D*), and in the ommatidium, before it has been separated into its component parts, have observed the passage of these fibers through the basal membrane.

The expanded proximal portions of the elongated pigment cells (Fig. 1 *B, p*) form the layer of pigment spoken of by Adensamer, as found on the basal membrane.

(7) Supplementary cells, four in number, are found at the same level as the cells of the inner tier. They are entirely different, and thus easily distinguished from the cells of the inner tier (*a*, Fig. 1 *D*).

(8) As shown in Fig. 2 *A* certain large nuclei were found in cross-sections at the distal part of the ommatidium. They are found only in the space between three ommatidia. They were not observed, nor the cells to which they belong, in maceration preparations.

BRYN MAWR COLLEGE, April, 1900.

REFERENCES.

- '80 GRENACHER. Ueber die Augen einiger Myriapoden. *Arch. f. mikr. Anat.* 1880.
- '93 ADENSAMER. Zur Kenntnis der Anatomie und Histologie von *Scutigera coleoptrata*. *Verh. d. Bot. Ges. Wien.* Bd. xliii.
- '96 ROSENSTADT. Zur morphologischen Beurtheilung der Augen von *Scutigera*. *Zool. Anzeiger.* Jahrg. xix. 1896.

BIOLOGICAL BULLETIN.

ABNORMALITIES IN THE CESTODE *MONIEZIA* EXPANSA. I.

IMPERFECT AND PARTIAL PROGLOTTIDS.

C. M. CHILD.

IN November, 1899, a large number of specimens of *Moniezia expansa*, a common parasite of the sheep, was obtained from the Union Stockyards in Chicago. Nearly every specimen exhibited one or more abnormalities in the form of the proglottids, but one specimen, some two feet in length, was found, which possessed over a hundred abnormal proglottids. This was incomplete, for in the oldest proglottids present the uterus had not yet appeared. The abnormalities in this individual are not different in kind from those found in others, but are much more abundant. Most of the cases described were selected from this specimen.

This paper, the first of a series, is devoted to a description of some of the simpler forms of abnormal proglottids. In the second paper a number of spiral anomalies will be described, and the third will include a general summary of the facts, together with some suggestions as to causes and significance.

The figures are all drawn with the aid of the camera from stained and mounted preparations. Figs. 2-6 are magnified about fifty diameters; all others about twenty. All except Figs. 7, 14, 15, 19, and 23 are taken from the single specimen mentioned above. These five figures are selected from different individuals.

Some of the figures show the dorsal side uppermost, others the ventral. The position is noted in most cases. In each figure the furrows of the lower side are drawn as broken lines. In cases where they gradually become shallower and disappear upon the surface of the proglottid, as they often do, the attempt is made to represent the general character of the line by a lighter or finer line in the figure on the upper side, and on the lower by longer spaces between the dashes composing the broken line. In the figures of abnormalities the testes are not represented. Nearly all figures show stages before the uterus appears.

The reproductive organs are represented schematically, for the exact details of structure are not essential to the object of this paper; but the position and relation of the organs is shown as exactly as possible.

Since it will be necessary to employ various terms with reference to the segments in the course of the description and discussion of the figures, it seems advisable, in order to avoid any possible confusion, to explain briefly the nomenclature employed. The terms "proglottid" and "segment" are used as synonyms; "anterior" and "posterior" possess of course the same significance as when applied to the whole animal; "transverse" is applied as referring to the direction perpendicular to the longitudinal axis of the animal, and parallel to the two flat surfaces, the ventral and dorsal; the "width" of a segment is equal to its transverse diameter; the term "longitudinal" refers to the direction parallel to the longitudinal axis of the animal and the "length" of a proglottid is equal to its longitudinal diameter. In the form under consideration the width of a proglottid is much greater than its length. The "thickness" of a segment is its dorso-ventral diameter. "Right" and "left" are used with reference to particular figures and do not always refer to right and left sides of the body. "Side" is used as referring to the region of the proglottid indicated by the preceding adjective, *e.g.*, "dorsal side," "right side," etc. The "inter-proglottidal furrow," "inter-segmental furrow" or "furrow" is the furrow or line which separates the proglottids. A "partial proglottid" is a portion of a proglottid incompletely or completely marked off by furrows. "Partial division" refers to the

incomplete separation of two proglottids or parts of proglottids, and the "partial furrow" is the furrow separating a partial proglottid from others; it may end free or may join another furrow. In many cases the furrows gradually become less and less distinctly marked and are said to become shallow as their depth is less than that of the normal furrow.

In anticipation of the summary it seems advisable to mention briefly some of the more important facts which may be gathered from the study of these abnormalities.

Taking as the basis for comparison the normal proglottid, numerous variations from this type are found. The segment may be longer or shorter than the normal, or may vary in length in different parts. The furrows bounding the segments may end at any point, leaving two or more segments partially united, or they may bend so as to run longitudinally. The furrows are evidently the expression of internal conditions, and where abnormalities in the furrows occur, the internal organs very often show abnormalities in arrangement and position which are very closely correlated with the position and development of the furrows. In brief, the position, development, and arrangement of the sexual organs are very closely correlated with the form and size of the proglottid. The organs which lie nearer the ventral side are affected chiefly by the form relations on that side, and those which lie nearer the dorsal side by the conditions there. This appears very clearly in many cases where the form relations on the two sides of the body do not correspond. Some cases appear to indicate that a certain degree of distinctness or separation, an "internal division," may exist without the appearance of distinct furrows. Between this condition and the normal, various degrees of division are indicated by shallower or deeper furrows. The various portions of the sexual organs, *e.g.*, the proximal and distal portions of the ducts, develop independently of each other *in situ*, and become connected secondarily, or in many cases remain separated. Abnormalities of the furrows are apparently due to the internal conditions in the growing regions. The abnormalities of the internal organs must be regarded as adaptations to the abnormal relations of form, size, etc., which already exist in the segment concerned.

Figure 1.

Before proceeding to the description of the abnormalities a brief description of the normal anatomy of the proglottid is necessary. In this species the proglottids are always much wider than long, but the relation between width and length varies considerably both with age and with the degree of contraction. Fig. 1 is a figure, viewed from the ventral surface, of a normal proglottid at the stage when the testes are ripe, or a little later, *i.e.*, about the stage when fertilization occurs. The furrows between the proglottids are schematically represented by a single line here, as in most of the figures. As a matter of fact, the posterior edges of the surfaces of each segment lap over the

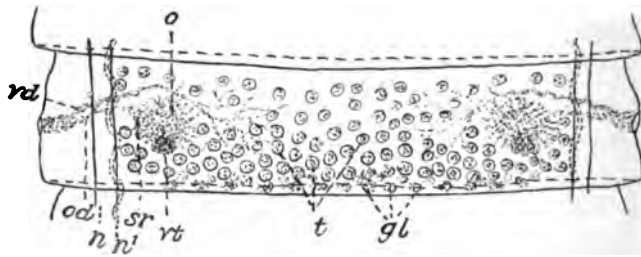


FIG. 1.

surfaces of the succeeding segment, *i.e.*, the furrow does not cut into the body perpendicularly to the surface but obliquely forward. Along the furrows on each surface, except near the edges of the segments, occur a varying number of small glands — the inter-proglottidal glands (Fig. 1, *gl.*), each of which opens by a distinct pore into the bottom of the furrow. At each side of the segment appear the two longitudinal nephridial tubes, the large ventral, *n*, and the smaller sinuous dorsal, *n'*. Transverse tubes, though visible in earlier stages, are very difficult to find later.

The terms “ventral” and “dorsal” are applied to the body of the cestode as follows: the ventral surface is the surface nearest which the ovaries and vitellaria lie, and the testes are situated near the dorsal surface.

Each proglottid possesses normally two pores lying near the middle of the two edges, but rather nearer the dorsal than the

ventral surface. The pore opens into the genital cloaca, or atrium, into which the male and female ducts also open. Following the female duct from this point, we find that it passes inward, somewhat anteriorly and dorsal to both of the nephridial tubes, then turns ventrally and posteriorly and opens into an enlarged portion, the seminal receptacle, *s.r.* Just beyond the seminal receptacle the ovary, *o.*, appears in the form of a rosette. The ovary consists of a mass of radiating branched tubules and is somewhat flattened in the same plane as the proglottid. The vitellarium, *vt.*, lies somewhat ventral and usually posterior to the ovary. At the stage shown in the figure the uterus does not appear, but in later stages it consists of an anastomosing set of tubes, which, after they receive the embryos, enlarge so as to fill nearly the whole proglottid. From this description it is seen that, although the ovary and vitellarium lie ventrally in the proglottid, the outer or distal portion of the oviduct lies dorsally. This point is important with regard to the relation of these organs in abnormal segments. Following the male duct from the atrium we find its terminal portion modified to form the cirrus. Beyond this the vas deferens, *v.d.*, follows the direction of the oviduct anterior to it, but is much coiled. It also runs dorsal to the nephridial tubes, but does not bend ventrally, as does the oviduct. Anterior to the middle region of the ovary it bends posteriorly and extends dorsal to the ovary toward the middle of the segment. Beyond the bend it begins to branch and soon breaks up into the fine tubules which connect with the testes. These latter, *t.*, lie scattered through the proglottid on the dorsal side, but are more numerous in the posterior half. They do not occur lateral to the nephridial tubes. Thus all of the male organs are nearer to the dorsal than to the ventral surface.

DESCRIPTION OF THE ABNORMALITIES.

All the figures except 7, 14, 15, 19, and 23 are taken from a single chain. These five are taken from as many different chains, and on comparing them with the other figures it becomes evident that the abnormalities found so abundantly in the one specimen do occur, though less frequently, in very many individuals.

The figures all represent cases of partial division of segments, together with the accompanying abnormalities in the form and position of the genital organs. A classification is difficult, and, I think, unnecessary. In general the more simple and regular cases are discussed first, the complex ones later. Cases resembling each other are grouped together as far as possible.

Figs. 2-6 are taken from various points near the anterior end of the chain. They all show stages before the appearance of the genital organs. These cases, although differing somewhat in form, are grouped together here as furnishing some evidence for the conclusion that the abnormalities of this kind appear at the time the furrows are formed and are not due to a later division of proglottids. They are certainly as common in these earlier stages as in later ones. Following these are grouped the cases in which the furrows on the two surfaces correspond closely. These include Figs. 7-15, as well as Figs. 2, 3, 5, and 6 of the preceding group. In Figs. 7-15 the genital organs, though they may be abnormal in position, are nearly always fully developed. The remaining figures, 16-23, show cases which are more complex and in which the furrows on the two surfaces do not usually correspond. Moreover, in these cases some of the genital organs are commonly rudimentary or abnormally developed.

Figure 2.

This figure was taken from the extreme anterior end of the body. The furrows between the proglottids have become fairly distinct. As the dorsal and ventral furrows correspond exactly in position, only one surface is represented in the figure. Four abnormal segments, *a*, *b*, *c*, and *d*, are present. The segments *a* and *b* are both examples of partial division, one upon the right side, the other on the left. Here

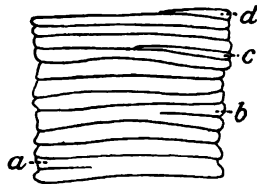


FIG. 2.

the partial furrows end free, not far from the middle of the segment in which they occur, so that the proglottid appears as if partially split from one edge. The two cases at the anterior

end, *c* and *d*, consist of partial segments on the right side. The separation of these is complete on both sides of the body.

Figure 3.

In this case two proglottids are incompletely separated on the upper side, because the furrow on the right side is slightly anterior to that on the left. The two parts of the furrow overlap slightly, *i.e.*, the left part extends past the inner end of the right part, and each ends free. On the lower side the furrow at the right bends anteriorly and meets the complete furrow anterior to it, thus marking off completely the small partial segment. The longer partial furrows correspond exactly on the two surfaces. Apparently the right and left portions of the furrows have been formed independently and have failed to meet.

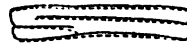


FIG. 3.

Figure 4.

In the segment *a* this figure shows a case where the segment is of less than normal length at the left edge, and where, moreover, the bounding furrows on the lower surface do not extend to the edge but bend so as to meet on the surface of the segment a short distance from the edge.

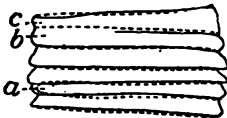


FIG. 4.

In *b* and *c* a rather simple case of partial division is represented. The partial furrow on the upper surface extends from the right edge to a point near the middle of the segment and there ends. At the left there is no furrow on the upper surface corresponding to this one, so the whole left half appears undivided. On the lower surface, however, the furrow between *b* and *c* is normal, extending across the whole body, meeting the upper furrow at the right edge and ending in a slight indentation on the left edge. At the right *c* is about twice as long as *b*, but at the left *b* is much the longer of the two. This difference is due to the fact that the furrows anterior to *c* are somewhat oblique.

Figure 5.

In this figure two variations from the normal form occur, both cases of incomplete separation or partial division. The partial segment *a* is completely separated on the upper surface from the segment in front, and its inner end is rounded, but on the lower surface the furrow between the two ends free, so that the separation is incomplete here. The segments *b* and *c* are

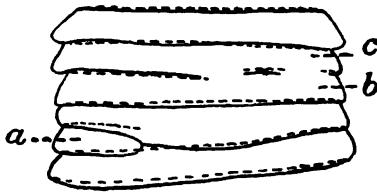


FIG. 5.

incompletely separated by four distinct partial furrows, all of which, however, lie in the same transverse plane and must be regarded as portions of a single furrow. The largest portion is at the left side, extending to the middle of the body on each

surface and of normal depth throughout. To the right of the middle a short partial furrow appears on each surface, the two being equal in length and in corresponding positions. At the right edge is a very short partial furrow marking off the two segments at the edge, but extending only a very short distance on either surface. The length on the two opposite surfaces of all the partial furrows, and especially of the two short entirely unconnected parts, is a point of interest. It is quite commonly, though by no means universally, the case that partial furrows, when they occur on the two surfaces, are of the same length on both.

Figure 6.

At the stage shown here the inter-proglottidal glands have begun to appear in the furrows between the segments. This case shows a rather unusual form of partial proglottid. The part *a* is completely marked off on both dorsal and ventral sides from the rest of the proglottid by the transverse furrow between *a* and *c*, and by the nearly longitudinal furrow between *a* and *b*, thus forming a small, distinct, partial proglottid. The transverse furrow extends somewhat beyond the point where the longitudinal line joins it, thus partially separating a small portion, *b*, from

the remainder of the proglottid. At *d* a triangular depression appears in consequence of the fact that the contours of the portions *a*, *b*, and *c* are somewhat rounded at this point where all three meet. The same conditions are present in some degree on the lower surface also, so that the thickness of the body at *d* is very slight. Inter-proglottidal glands appear just anterior to the extra transverse furrow, thus showing the same relation to it as to the normal complete furrow. This is usually the case, provided the abnormal furrow attains a certain degree of depth.



FIG. 6.

Figure 7.

This is a case of partial division seen from the ventral side. The segment is undivided at the left, and one genital mass appears. To the right of the middle, however, a short partial furrow appears on each surface, the two corresponding closely in position. The right edge shows clearly a division into two segments, and a very short furrow extends from it on to the ventral surface. In accordance with these indications of division in the right half two genital masses appear.

The fact that the two partial furrows correspond so closely on the two surfaces indicates that they are distinctly the result of internal conditions. Judging from the existence of the two genital masses and the short furrow at the right edge, it appears probable that division or separation exists in a certain degree between the two regions, even where the actual furrows do not appear.

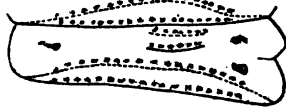


FIG. 7.

Many other cases support this view. It would appear that the individuality of the segment must attain a certain degree of development in order to cause the formation of furrows, and that, where only partial furrows exist, the division may be in many cases more complete than the furrows indicate.

The short furrows on the two surfaces bear inter-proglottidal glands like the complete furrows.

Figure 8.

This case, though at a later stage of development than Fig. 7, resembles it. Two partial furrows appear, one on the dorsal, the other on the ventral surface, corresponding exactly in position and length, but entirely unconnected. The exact correspondence in position and length of these two entirely unconnected furrows indicates very clearly, as does the similar condition in Fig. 7, that



FIG. 8.

the position of the furrows is determined by internal conditions, for it is difficult to understand how two perfectly similar partial furrows could arise on opposite surfaces of the body except

as the expression of certain internal form-producing conditions.

The genital organs are duplicated on the left side, but the two pores are approximated. The individuality of the two portions is apparently not sufficient to give rise to furrows at the edge, so that pores tend to appear near the middle of the edge. But the furrows extend almost to the edge, and the existence of two pores is undoubtedly the result of this position. The right side shows no trace of duplication.

Figure 9.

This figure shows three cases of partial division, and in all the partial furrows end free and correspond

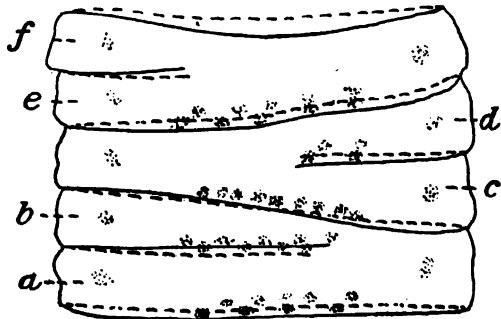


FIG. 9.

on the two surfaces. The partial furrows between *a* and *b* are most nearly complete, extending past the middle of the body. Between *c* and *d* they are shorter, and between *e* and *f* still shorter. In each case the two partial segments are longer than the corresponding single segment at the opposite edge, consequently oblique furrows appear between the three sets, but,

owing to the alternation in position of the partially divided regions, the length of the right and left edges of the whole group is the same, *i.e.*, the abnormality in form of each segment compensates for that of the others. Each partial proglottid possesses its own genital mass, so that there are five on the left side of the group and four on the right. All partial furrows that are long enough, *i.e.*, all except that between *e* and *f*, show interproglottidal glands, and all are of normal depth and distinctness. The decreasing length of the partial furrows from the posterior to the anterior set is noticeable, but whether it possesses any special significance or not is not clear.

Figure 10.

Four cases of partial division of proglottids occur here, three on the left and one on the right (*a*, *b*, *c*, *d*). In each case the partial furrows on the two surfaces correspond almost perfectly in position and length and end free, not far from the middle of the body. The relations at the edges do not differ from the normal except in the case of *b* at the right. This proglottid is partially divided at the left, but the undivided portion to the right is as long as the two partial proglottids at the left, though no furrows appear. The complete genital mass *g* appears at the normal distance from the furrows bounding the segment posteriorly, and

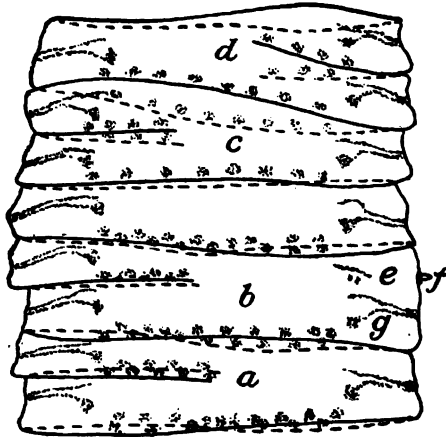


FIG. 10.

a partial second set of organs, *e*, *f*, appears in the anterior region, consisting of the inner portion of the vas deferens, *e*, two small groups of cells just posterior to it, which probably represent the inner portion of the oviduct or the ovary, and entirely unconnected with these at the right edge a pore with

protruded cirrus, *f*, which has probably been forced out through the pore by the pressure during fixation. Here then is a complete set of genital organs and a second partial set occurring without any furrows between them. This condition is rare. I have found only one other similar case.

This appears to be a duplication of genital organs in a proglottid which is morphologically single, at least in its right half. I believe, however, that this case is simply another example of the fact that a certain degree of individuality may exist without the appearance of furrows, but may still be quite sufficient to lead to the partial formation of genital organs. The fact that the proglottid is divided on the left side into two parts by partial furrows of normal depth affords additional evidence for this view. It is evident that the causes leading to the formation of genital organs at *e* and *f* is much less efficient than normally, for the organs are extremely rudimentary and can never function in the normal manner.

The presence of furrows on the surface is, in general, simply the morphological expression of certain internal conditions. These relations differ in degree in different species, and, as is evident from the variations discussed in this paper, in this species also. This being the case, the logical conclusion seems to be that a certain degree of isolation or individuality may exist without the appearance of furrows on the surface.

In this rudimentary and incomplete set of organs, *e* and *f*, it is seen that the two parts, *e* and *f*, arise independently of each other. The pore and cirrus are absolutely unconnected with the inner portions of the ducts which are present. These facts show that the proximal and distal portions of the genital organs arise independently *in situ*, in, or as nearly as possible in, the position which is normal for each.

The cells of the incomplete set show the same degree of differentiation and the same reaction to the stain as the corresponding regions of the complete set, *g*. It seems probable, therefore, that both sets were formed at the same time. The differences between the two sets consist in the entire absence from the one of certain parts present in the other. As will appear below, a segment of less than normal length usually possesses only partial

genital organs. Here no actual furrows are present, but the relative positions of the two sets of organs, *g* and *e f*, indicate that the set *g* corresponds to a longer portion of *b* than does the set *e f*.

Inter-proglottidal glands appear in all of the partial furrows.

Figure 11.

This figure, representing a case of partial division, shows very distinctly an almost complete gradation in individuality from the right to the left, as indicated by the arrangement of the organs. At the right the partial furrows separate the segments in a normal manner, but the posterior segment is the shorter. Correspond-

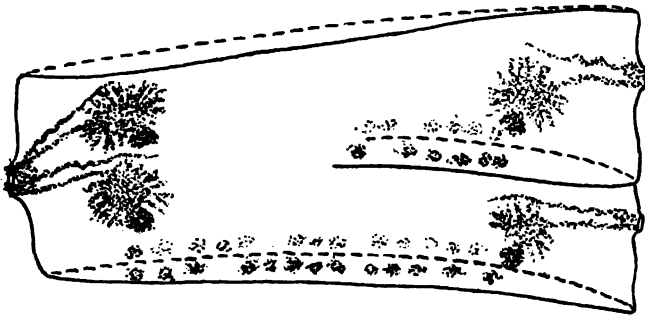


FIG. 11.

ing to the position of the furrows, two complete sets of organs appear on this side, but the posterior set, especially the ovary, is smaller and less fully developed than the anterior.

The partial furrows end, however, near the middle of the body, leaving the left half apparently undivided, and the left edge is considerably shorter than the right. That the two segments possess a certain degree of individuality beyond the region where the furrows end is indicated by the presence of two complete sets of organs and ducts, which are, however, closely approximated and open through a single pore situated at the middle of the undivided edge. Even the terminal portions of the two sets of ducts are distinct and two cirri are present. To judge from the arrangement of the organs it appears that from right to left the segments are less and less completely separated, until at the left edge the

conditions are nearly those of a single normal segment, so that only a single pore appears.

Inter-proglottidal glands lie in the partial furrows on each surface.

Figure 12.

The figure, a view from dorsal surface, shows three segments which are all incompletely separated at the right side. At the left the separation is complete, the furrows appear normal, and the genital organs in process of formation are normal in position and form. On the right the furrows separating the segments *a* and *b* end at *d* and *e*, before reaching the edge, the furrow on the ventral side becoming shallow and rather irregular in its

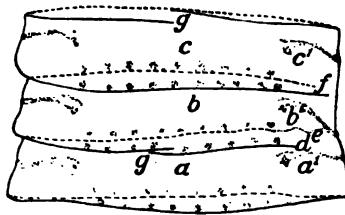


FIG. 12.

course (*e*) but extending almost to the edge, while the dorsal furrow ends more abruptly at a greater distance from the edge (*d*). The furrows between the segments *b* and *c* do not reach the right edge, but end rather abruptly near it at *f*, the points of

termination on the two surfaces of the body being about equidistant from the edge. Thus the whole right edge shows no traces of division into separate segments, but nevertheless possesses three genital pores, two of which are near together. The ovaries and vitellaria and the inner ends of the vasa deferentia at the right of *a* and *b* are normally situated with regard to the furrows, for at this distance from the edge the relations are practically those of two normally distinct proglottids. As we approach the right edge, however, the dorsal furrow, *d*, ends abruptly at some distance from the edge, while the ventral furrow, *e*, becomes more shallow and finally disappears near the edge. The terminal portions of the ducts lying nearer the dorsal surface are affected in greater degree by the relations on the dorsal surface, and we find here that as the ducts approach the edge they also approach each other, the approximation being almost wholly due to the abnormal direction of the ducts of the set *b'*. The organs at *a'* lie in the normal position, but those at *b'* lie

obliquely in their segment, the ducts extending outward and posteriorly toward the pore; but these positions are, I believe, due to the relations of the segments to each other. The fact must be recognized that in segments of normal length a complete set of organs capable of functioning tends to form, however abnormal its position; *i.e.*, the parts are formed independently and tend to unite in the normal manner. In this case the proximal portions of the organs *a'* and *b'* are formed in their normal position with respect to the furrows bounding the segment ventrally. The edge of *a b* shows no dividing furrow, so that it might be expected that a pore would appear at its middle. The furrows *d* and *e* approach near the edge, however, and the existence of a certain degree of "internal division" at the edge is probable. Thus two pores are formed instead of one, but are separated by less than the normal distance between pores of two successive segments. Apparently the degree of separation between the two segments at the edge is only slight, so that the edge is more or less like that of a single segment, and the middle region is the pore-forming region. But the two segments are sufficiently independent to give rise to two pores instead of one common to both; and these two pores, it will be noticed, are equidistant from the middle of the undivided edge of *a b*. But the pore in *b* is far posterior to the ovary, etc., and in order that the two may be connected the ducts must extend obliquely, as they do. In *a*, on the other hand, the pore is directly lateral to the proximal portions, and thus the ducts are horizontal.

The furrows between *b* and *c* extend almost to the edge, so that conditions here approach very closely to the normal. The organs *c'* in *c* are normally situated as if the furrow *f* were complete.

Two of the furrows on the dorsal surface are interrupted (*g g*) and the two parts overlap slightly in each case.

Figure 13.

The variation from the normal form shown here is almost identical with that shown in Fig. 12, *a* and *b*, except that here the two partial furrows bend anteriorly at the right. Both become

shallow and indistinct and terminate on the surface near the right edge. As in Fig. 12, *a* and *b*, two complete sets of genital organs appear on the right side, the posterior set being normal and the anterior set situated somewhat obliquely, with the pore

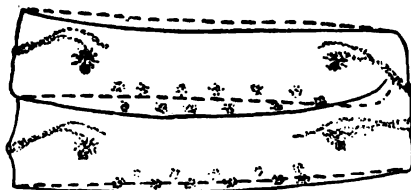


FIG. 13.

near that of the posterior set. The positions of the two sets of organs are undoubtedly the result of conditions similar to those in Fig. 12, *a* and *b*, and are to be explained in the same way.

It is noticeable that the curve at the right ends of the furrows appears to have no significance as regards the position of the organs, which are situated as they would be if the furrows ended without bending forward. The furrows become very slight here, being little more than wrinkles on the surface.

Figure 14.

Two cases of partial division, *a b* and *c d*, are shown in this figure, viewed from the ventral surface. The partial furrows on the two surfaces correspond in both cases. Between *a* and *b*

they extend from the left edge to a point just beyond the middle of the body, thus leaving almost the right half undivided, and, corresponding to the partial division, one set of organs is found

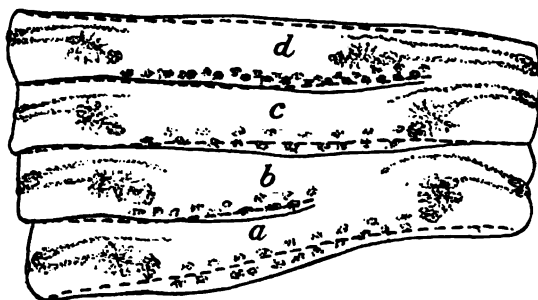


FIG. 14.

at the right, while two appear at the left. The division between *c* and *d* is more complete, extending from the left edge over about three-quarters of the width of the body, and in this case, although the right edge itself shows no furrow, two sets of organs occur at the right as well as at the left, but their pores are much

nearer together than at the left, where the separation of the segments is complete and normal. It appears from the arrangement of the organs at the right that the two segments *c* and *d* do preserve a certain degree of individuality in the region beyond the end of the partial furrows, for if this were not the case we should expect to find only one pore instead of two, a condition which does frequently occur. The position of the ovary and vitellarium at the right of *d* is peculiar. The ducts, instead of bending posteriorly, extend straight inward, and the ovary lies nearer the middle of the segment than the others. This position is apparently due to the form relations here. The distal portions of the two sets of organs at the right of *c* and *d* are approximated, but the partial furrows between *c* and *d* extend to the region of the ovaries, and *d* is slightly shorter in this region than *c*, so that the ovary and vitellarium in *d* take the position in which they can attain most nearly their normal development.

Inter-proglottidal glands occur in the partial furrows as well as in those which are complete.

Figure 15.

This figure shows a case of partial division in which the partial furrows are nearly complete. The segments are rather old, the ovaries and ducts being in process of degeneration and the uterus containing embryos (not drawn in figure).

At the left side the segments are normally bounded, and the genital organs are apparently normal. At the right the partial

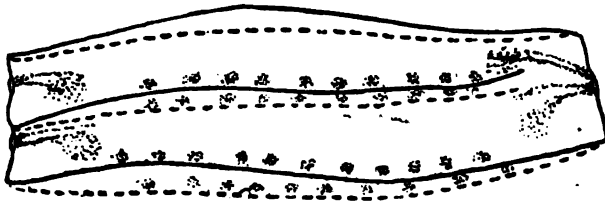


FIG. 15.

furrows end before reaching the edge, and two sets of organs occur, opening into a common pore. The partial furrows extend so nearly to the edge that in the ovarian region the two segments

appear almost normally separated, and accordingly two sets of organs appear. The edge, however, is undivided and shorter than the combined length of the two segments at the left, and only a single pore is formed, into which both sets open. This case shows much the same gradation from complete division to almost complete union, as is found in Fig. 11, but here the gradation occurs within a much shorter distance, for the furrows are nearly complete, while in Fig. 11 they extend only halfway across the body.

Figure 16.

The figure represents a peculiar case of partial division seen from the dorsal side. The two segments are completely separated at the left by partial furrows which extend to the middle on each surface. Corresponding to this separation we find two

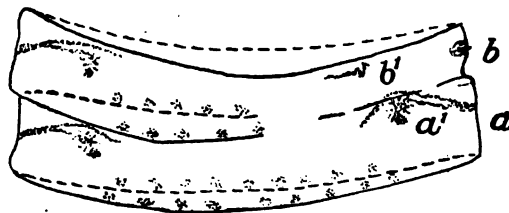


FIG. 16.

complete sets of genital organs. On the right the two portions are separated dorsally by a very shallow furrow which is interrupted at several points.

This furrow passes the right edge and extends for a very short distance on the ventral surface, leaving all the rest of the right half of the ventral surface undivided. Corresponding to this incomplete separation we find abnormalities in the genital organs. There is one complete set of organs (a') in the posterior segment, and portions of a second set (b') in the anterior segment.

The ovary, vitellarium, and inner end of the vas deferens of a' lie nearly in the middle of the undivided ventral side, almost directly beneath the shallow interrupted furrow on the dorsal surface, thus indicating that their position is more directly affected by the conditions of the surface to which they are nearest, *viz.*, the ventral. The pore and atrium, on the other hand, appear on the edge of a , nearer the anterior than the posterior end. The partial furrow on the dorsal surface and the right edge is very slight, *i.e.*, the two partial segments, though

separated to a certain degree on this side, are still much less distinct than normal segments, and the pores and terminal portions of the ducts of *a'* are therefore found in a position only a little posterior to that which they would occupy if *a* and *b* were not separated at all.

The partial segment *b*, as marked off by the slight and interrupted dorsal furrow, is narrower than *a*. The complete set of organs, *a'*, lies nearly in the middle of *a b*, but since the two segments are sufficiently distinct dorsally for the formation of a slight furrow, there is a certain tendency for another set of those organs which lie near the dorsal surface to form in *b*, as is indicated by the presence of a pore with atrium and cirrus at the edge, and the inner portion of the vas deferens at *b'*. These parts are wholly unconnected, and there is no trace of female organs. The failure to develop a complete vas deferens is probably due to the fact that the segment *b*, as bounded dorsally, is of less than normal length and imperfectly separated from *a*. The parts of the male organs which do appear are identical with those which we find in Fig. 10, *b*, but in that case there is a trace of female organs also. These two cases are examples of a condition often found in short, imperfectly separated segments, *viz.*, the development of the innermost and outermost portions of the organs without connection. It appears as if the region of the pore and the inner portions of the ducts represent, as it were, the places of least resistance with respect to the formation of the genital organs, so that segments which are not sufficiently normal to give rise to a complete functional set of organs may form these two parts, but not the ducts connecting them.

The complete absence of female organs at the right of *b* shows very clearly that the formation of the ovary and vitellarium is connected with the conditions on the ventral side of the body, and the formation of the vas deferens with conditions on the dorsal side.

The shallow dorsal interrupted furrow at the right is without inter-proglottidal glands, probably because of its slight development.

Figure 17.

The figure represents two partially separated segments seen from the dorsal surface. At the right the division is complete and the genital organs are normal.

The ventral furrow extends without interruption about three-quarters across the ventral surface, but is somewhat irregular in its course. The dorsal furrow is interrupted at two points, once just to the right of the middle and again near the left side, leaving a short portion, *c*, entirely separated from the rest. This portion does not reach the left edge, but turns anteriorly near it and ends abruptly. The ventral furrow shows no portion corresponding to this portion, *c*, consequently the segments are



FIG. 17.

visibly separated only on the dorsal surface in this region. The abnormal relations of the furrows are accompanied by abnormal conditions in the genital organs. The case is somewhat similar to the one

appearing in Fig. 16, and confirms the conclusions reached in the discussion of that case. The posterior portion, *a*, is longer than *b* and possesses a normal set of organs normally situated, while in *b* there is a complete vas deferens and pore, but no trace of ovary, vitellarium, or oviduct. Analysis shows a very close relation between the organs and furrows. The case is very similar to Fig. 16, but presents some differences. The partial furrow *c* is dorsal, but is deeper than the furrow in Fig. 16; *i.e.*, the division between *a* and *b* is a little more complete here than there, and the ovary and vitellarium of *a* lie further posteriorly. A second set of female organs does not appear, probably because the region *b* is not separated from *a* on the ventral side and is considerably shorter than *a*, so that the single ovary and vitellarium serves for the whole length of *a b*. On the dorsal surface the partial furrow *c* shows that different relations exist, and here in the shorter portion, *b*, there is formed a complete vas deferens and pore. The pores in *a* and *b* are

approximated, this being apparently due to the fact that the division is incomplete and the furrow *c* does not reach the extreme edge. Comparison of this figure with Fig. 16 is very instructive. The relations of the furrows at the left of Fig. 17 are almost the same as at the right of Fig. 16, the chief visible differences being that in Fig. 16 the furrow is shallower, but passes over the edge, while in Fig. 17 it is of normal depth but does not extend to the edge. In both cases the corresponding portions of the ventral surfaces are without furrows. As regards the genital organs in the shorter anterior portion *b* in the two cases, we find in Fig. 16, where the dorsal furrow is shallow, only the inner portion of the vas deferens and the pore appear, the two being entirely unconnected, while in Fig. 17, where the dorsal furrow is deeper and thus more nearly normal, a complete vas deferens is formed connected with its pore. Moreover, in Fig. 17 the furrow does not reach the edge, and the pore in it is situated somewhat posteriorly, while in Fig. 16, where the furrow passes the edge, the pore is in the middle of the edge of *b*. In neither case does the region *b* show any trace of female organs. The conclusions regarding the causes of the conditions in Fig. 16 apply with equal force here. To my mind these two cases afford ample basis for the views expressed in this paper, but these are supported and confirmed by a mass of evidence from the other abnormalities discussed here, so that the conclusions reached become not only probable, but, I believe, incontestable.

Figure 18.

In this case a small partial segment is completely marked off by very slight furrows, and the anterior furrows show a very abrupt bend where the partial segment ends. In the complete segment lying just posterior the genital organs are normal, but in the small partial segment nothing but two small groups of cells appear, and it is impossible to determine just what portion of the organs they represent. The pore is entirely absent.

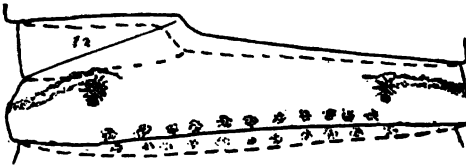


FIG. 18.

Figure 19.

In these segments the ovaries and ducts are degenerating, and the embryos are in the uterus (not shown in figure). The two segments seen from the ventral surface are incompletely separated ventrally by a partial furrow which does not reach the left edge. Dorsally the furrow between the two is complete. At the right the segments and genital organs are normal. At the left *a* is longer than *b* and is separated from it only dorsally. In *a* a complete set of organs appears, but situated somewhat farther anteriorly than the normal position, *i.e.*, approaching

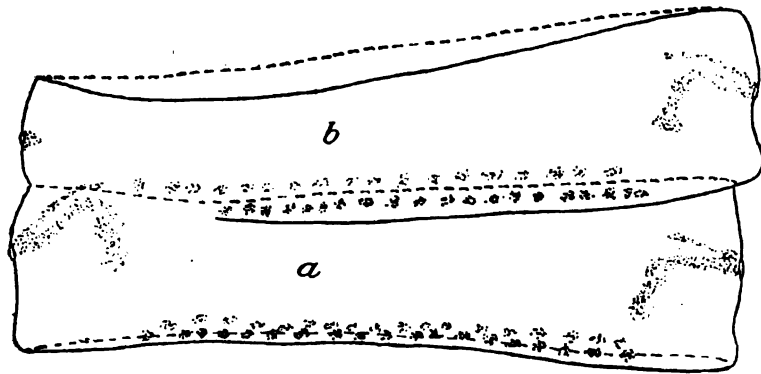


FIG. 19.

the middle of the undivided region of *a b*. The dorsal boundary between *a* and *b* is normal at the left, and the edge is divided, and in *a* we find a pore almost normally situated with respect to the form of the dorsal surface. In the shorter segment *b* the only indication of genital organs at the left is a pore, but this is placed nearly in the middle of the edge of *b*, *i.e.*, almost normally with respect to the dorsal boundaries. The fact that no other organs appear here is doubtless due to the shortness of this portion of the segment *b* and to its imperfect separation from *a*. As seen in Figs. 10, *b*, 16, 17, and 18, imperfectly separated segments of less than normal length usually possess more or less rudimentary organs. As noted, the two pores at the left of *a* and *b* are not quite in their normal positions, *i.e.*, they are separated by less than the normal distance, as is evident from a

comparison with the pores at the right. This approximation of the pores is evidently due to the incomplete separation of the two segments, which, though separated dorsally, are united ventrally. Thus, while the existence of the pores seems to be determined by the relations upon the dorsal surface, their position may be affected in some slight degree by the relations on the ventral surface.

Figure 20.

The series of abnormalities shown here is rather complex and may be considered most conveniently segment by segment. The ventral surface is uppermost.

The segment *a* is partially divided on the left side by a furrow which extends from a point on the ventral surface very near the edge around the edge to the dorsal surface, and for a short distance on the dorsal surface, where it ends free. The degree of separation is sufficient to cause the appearance of two distinct and complete sets of genital organs. At the edge the division is complete, and accordingly the pores lie in practically their normal positions on each side of it. The division, as indicated by the furrow, extends only a short distance on either surface, but farther on the dorsal than on the ventral surface, and the arrangement of the ducts, ovaries, etc., is in accord with these relations. The two ovaries, etc., are quite closely approximated, but the ducts diverge, thus indicating that the division becomes more complete with the approach of the edge. This case, like Fig. 10, *b* and *c*, illustrates, though in a less degree, the apparent existence of a certain degree of separation in regions where the furrows do not appear. Thus there is no furrow on either side immediately between the ovaries, yet two sets appear. That the division is more complete dorsally than ventrally is shown by the fact that the dorsal furrow is longer than the ventral, and the position of the genital organs accords with this fact as shown above. At the right *a* shows no trace of division, and a single normal set of organs is present.

The segments *b* and *c* are best considered together. At the right, before reaching the edge, both the dorsal and ventral furrows separating *b* and *c* bend anteriorly and become very slight,

being mere wrinkles on the surface. The ventral furrow can be traced to the anterior boundary of *c*, where it ends very near the edge, while the dorsal furrow ends free about midway of the segment. Thus the segment *c*, as bounded by the very shallow curved furrows, does not reach the right edge of the body at all on the ventral surface, and dorsally extends to the edge only anterior to the end of the curved furrow. The portion forming the edge is not separated from *b*. At the left the ventral furrow

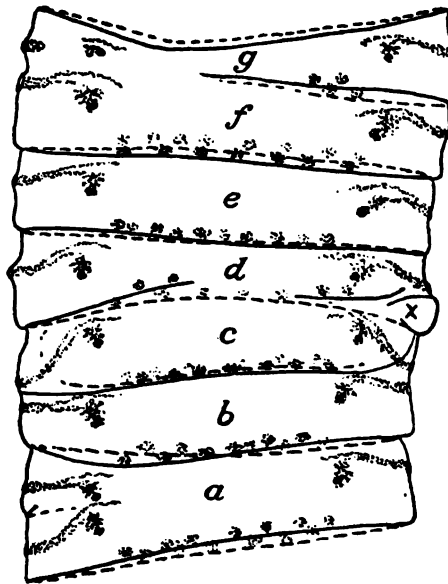


FIG. 20.

between *b* and *c* reaches the edge normally, but ends there, for the dorsal furrow, instead of extending to the edge and meeting the ventral, bends forward like the right ends of the furrows and becomes like them very shallow, a mere wrinkle, which is visible to the anterior end of the segment. Thus the portion forming the left edge in the region of *c* is connected on the ventral surface with *c*, but dorsally only with *b*, *i.e.*, there is a short spiral here. This case shows how a spiral may arise by the bending forward of one of the furrows to meet the next in front, instead of uniting with its fellow on the opposite surface. In nearly every case of spiral variation this bend occurs as it does here near the edge, though in many cases the furrow remains of normal depth. This case then is what may be called an incipient spiral modification.

The spiral does not appear between *c* and *b* at the right, simply because both furrows bend forward, though the furrow does not completely separate the two segments, but ends free. The furrows anterior to *c* must be considered briefly before turning to the discussion of the genital organs of *b* and *c*. The dorsal

furrow is complete and takes a slightly curved course, but the ventral is divided into three parts. At the left one portion extends from the edge obliquely inward and anteriorly to a point near the middle, where it ends. The second portion lies on the right side, is almost transverse, and extends nearly to the right edge, overlapping with the third portion which reaches the edge at a point anterior to the corresponding dorsal furrow. Upon the edge it turns posteriorly and passes backward to the point where the dorsal furrow reaches the edge, and there it turns inward again, extends for a short distance, and terminates freely, thus almost surrounding a small region on the ventral surface at *x*. Notwithstanding these irregularities the furrows between *c* and *d* approximate to the normal conditions, but the ventral furrow is a little anterior to the dorsal, except in the short portion posterior to *x*. At the right relations are normal.

Returning now to the genital organs in the segments *b* and *c*, we find in each segment a complete set on each side. Considering first the organs in the left side of *b* and *c*, it is seen that in *b* the inner portions are normally placed, but the pore lies rather more anteriorly than its normal position. As noted above, there is a short spiral here owing to the course of the dorsal furrow in *c*, and the edge corresponding to *c* is not separated dorsally from *b*. This accounts for the position of the pore in *b*; *i.e.*, the organs in *c* are situated very much as they would be if there were no dorsal furrow at all between *b* and *c* in this region. The only indication of division on the dorsal surface in the region of the ducts is the very slight furrow curving forward. This seems not to affect the course of the ducts at all, for they cross it at right angles to reach the edge. The extreme posterior position of the pore in *c*, *i.e.*, its approximation to that of *b*, is evidently due to the same causes as the displacement of the pore in *b*, *vis.*, the absence of dorsal division in this region.

At the right in *b* and *c* somewhat similar conditions exist. The positions of the inner portions of the organs are about normal. As regards ducts and pores on the right, there is the same approximation as on the left. This end of the dorsal furrow in *c* is incomplete, ending, after bending forward, free on the surface just dorsal to the middle region of the ducts, and the ventral fur-

row also bends forward. The ducts in *c*, however, are situated as if the curved portions of the furrows were not present; that is, as if the furrows, especially the dorsal, ended about where they begin to bend.

The right edge corresponding to *b* and *c* is undivided, and the pores are accordingly approximated as on the left side. The separation of the two segments is normal up to within a short distance of the edge, and thus probably determines the existence of two separate pores, instead of the union of the two sets of organs in one.

The significance of the curved ends of the furrows in *c* requires a brief consideration. As mentioned above, they are very slight, being mere wrinkles, and though they are continuous with the normal inter-proglottidal furrows, they are not like these in appearance. The position of the genital organs does not appear to bear any direct relation to them, for the ducts cross them to reach the edge. Undoubtedly the slight development of these curved ends indicates a very incomplete separation of the parts which they bound, and, as will appear later, it is possible that such furrows do not always coincide with the real segmental boundaries.

The position of the organs in *c* and *d* appears to be nearly normal. At the right the pore lies very near the end of the abnormal ventral furrow, but about in the middle of the edge, as bounded dorsally, thus showing that its position is determined, at least largely, by the relations on the dorsal side. At the left the pore is approximate to the pore in *b*, evidently because of the absence of division on the dorsal surface at the edge.

The segment *e* is apparently perfectly normal, but *f* and *g* show abnormal relations, being separated on the right but united on the left. The partial furrows extend from the right edge a short distance past the middle of each surface and end free, the terminal portions being shallower than the rest. Thus the left side is without any true furrows, but the surface shows certain indications of a division between the two parts. From the end of the furrows to the edge there extends a depression in each surface too broad and indistinct to be called a furrow, but still apparently indicating a certain degree of separation (not shown in the figure). It reaches the edge in the slight depression between the two

pores at the left of the figure. The existence of this line of depression indicates, I believe, that *f* and *g* are really more or less distinct segments, even on the left, where no true furrow occurs. At the right *g* is as long as *f*, but is shorter at the left, and the furrows bounding it anteriorly are abnormal, for they are not transverse but extend from each edge somewhat posteriorly, thus making *g* very narrow just to the left of the middle. At the right the genital organs in *f* and *g* are normal and normally placed. At the left the organs in *f* are situated a little anterior to their normal position. We find, however, a second partial set of organs anterior to the first and showing relations almost identical with the rudimentary organs in *b* in Figs. 10, 16, and 17. The inner portion of the vas deferens appears, and just posterior to it lie small groups of cells which apparently represent a portion of the female organs. These parts are, however, entirely unconnected with the cirrus and pore, which are of normal size and appearance. Here again the inner and the outer portions have developed independently of each other, and the connecting ducts are absent. This case differs from those in Figs. 16 and 17, and resembles that in Fig. 10, *b*, in that a portion of the female organs appears here. In Figs. 16, *b*, and 17, *b*, a distinct furrow occurs on the dorsal side, but there are none ventrally; this condition indicating a more complete separation on the dorsal side than on the ventral, while in Fig. 10, *b*, as in this case, distinct furrows are absent on both sides in the immediate region concerned. Probably portions of both female and male organs occur in these cases, because the degree of separation, though slight, is the same on both sides. The incompleteness of the organs is doubtless due here, as in the other cases, to the small size of the segment.

The inter-proglottidal glands appear on all the furrows which lie within the zone of their formation. In the furrows between *f* and *g*, however, they are found only near the right side. The terminal portions of the furrows are shallow, and thus apparently insufficient to cause the glands to appear.

The region from which this figure was taken is not exceptionally abnormal, but was selected because it presents a number of different kinds of abnormalities near together.

Figure 21.

The figure represents four incompletely separated proglottids seen from the ventral surface. Between *a* and *b* the furrows at the left meet at the edge and extend over about one-third of each surface, ending free. At the right a peculiar curved furrow appears on each surface, and on the dorsal surface an oblique furrow extends posteriorly over *a* from the curved position almost to the posterior boundary of the segment. In accordance with the relation of the furrows, the genital organs at the left are

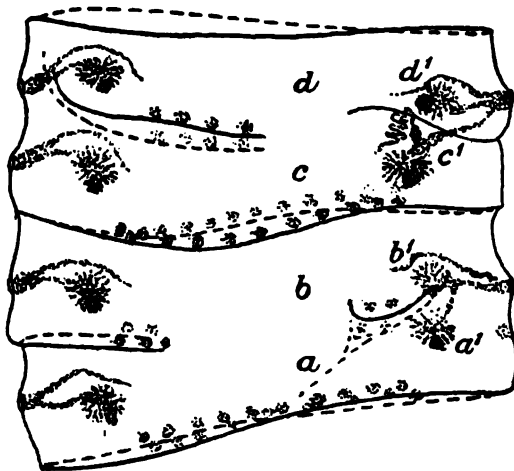


FIG. 21.

normal, but on the right side of *a b* a peculiar set of abnormalities appears. Two distinct ovaries and vitellaria, *a'* and *b'*, appear, one in *a*, the other in *b*, but there is only one vas deferens, that in *b*. The oviduct from *a'* extends anteriorly and unites with the oviduct of *b'* near the seminal receptacle, and the com-

mon duct opens through the pore in *b*. In a number of cases I have found two sets of organs opening through a common pore, but the union of oviducts at a point so far from the pore is rare. The organs *a'* consist wholly of female organs, the vas deferens being entirely absent. It will be noted that the partial furrow on the dorsal surface in this region takes an oblique course. The right end of the curved partial furrow on the ventral surface takes a similar course. The ovary *a'* lies quite near these furrows and consequently there is no space for the development of the vas deferens in anything like its normal position. As the relative position of the various organs appears to be quite definitely determined in all cases, there is probably no tendency

for the vas deferens to appear at all when it cannot be formed somewhere near its normal position. The partial furrows end just between the ovaries of a' and b' , and accordingly the two are quite closely approximated. This may be another reason why the vas deferens does not appear in a' , and it is, I believe, because of this approximation that the oviduct of a' unites with that of b' instead of running independently to the surface. There is a small rudimentary pore on the right edge of a which is wholly unconnected with the ovary a' , thus showing again the tendency for the terminal portions of the genital organs to develop separately in the segments of a low degree of individuality. The genital organs, b' , appear normal in form and constitution. The vas deferens is present here and in its normal relations. The pores in a and b are somewhat approximated, owing, apparently, to the fact that the edge is undivided, though division exists a short distance from it.

Between c and d at the right there is a distinct, though rather slight, furrow only on the ventral surface, and this furrow becomes somewhat oblique a short distance from the edge, *i.e.*, it extends somewhat anteriorly from the edge toward the middle. Its slight development is not clearly shown in the figure, but it is much less deep than the normal furrow. Dorsally there is no distinct furrow but only a shallow depression extending in the same direction as the furrow on the ventral surface and terminating in the corresponding region (not shown in the figure). As in the case of Fig. 20, f and g , I believe this depression represents what might be called a very rudimentary furrow, and its correspondence in this case with a distinct furrow on the ventral surface supports this view as regards both this case and Fig. 20. The genital organs, c' and d' , show peculiar relations. There are two complete sets opening by a common pore in d' . In c' , however, the vas deferens does not extend to the pore at all, but forms a complex coil just anterior to the ovary and *apparently opens directly into the seminal receptacle*. A small cirrus appears to be present in the enlarged terminal portion of the male duct which is seen in the figure. This is the only case where such a relation of the male and female organs has been found, unless it occurs on the left side of e , Fig. 22, where

it is impossible to determine the exact relations. In the organs at *c'* the portion of the seminal receptacle which lies between the ovary and the point of union of the male and female ducts is full of spermatozoa, while the outer portion is entirely empty, and this fact renders it certain that an actual union of the ducts with an opening does occur, and shows, too, that self-fertilization occurs as well.

It is important to note that here in *c* a vas deferens is formed, while in *a*, as mentioned above, the male duct is absent. The cause of this difference is indicated by the different direction of the partial furrows in the two cases. The partial furrows between *a* and *b* at the right extend obliquely backward posteriorly from without inward, and cut off from the segment *a* the region where the vas deferens would normally form, while the ventral furrow and the dorsal depression between *c* and *d* slant anteriorly, and thus a space is left in *c* anterior to the ovary where the vas deferens may form. The fact that the vas deferens opens into the seminal receptacle instead of extending to the pore, while the oviduct pursues a more nearly normal course, is perhaps not explicable on the basis of the form relations of the segments, but the suggestion offers itself that the close approximation of the two sets of organs, *c* and *d*, prevents its formation between them where it would naturally appear; so that it is confined wholly to the inner genital region. The oviduct of *c'* crosses the furrow to reach the pore instead of opening in its own segment. This is probably due to the fact that the degree of separation between the two segments is less than normal; for, as mentioned above, there is no distinct furrow on the dorsal surface, though ventrally the segments are sufficiently separated to give rise to separate ovaries. No pore at all is found at the right edge of *c*, and this is probably due to the same fact, *viz.*, that dorsally the degree of division is very slight, so that only one pore is formed for the two segments, though the position of this is apparently affected in some degree by the partial division which does exist, since it is placed somewhat anterior to the middle of the edge of *c d*.

In the genital organs *d'* of *d* relations are almost normal, but the oviduct is shorter than in *c'*, and the ovary is thus nearer the edge of the proglottid. The oblique direction of the partial fur-

row may perhaps account for this difference in position in the two sets of organs, for the line connecting the centers of the two ovaries lies at right angles to the furrow separating them, and the ovary of *d'* thus lies in a region where the segment *d* is longer than it is in the region where the ovary would normally appear.

The relation of the furrows and genital organs at the left of *c* and *d* requires little comment. The organs are normal in all respects. The two partial furrows approach close to the edge, but turn forward, becoming very slight, and soon disappear. The pores are somewhat less than the normal distance apart, for the furrows do not quite reach the edge. The curve forward of the outer ends of the furrows apparently does not affect the position of the genital organs, this portion of the furrows being very shallow.

Inter-proglottidal glands appear in all the partial furrows except the one between *c* and *d* on the right, and their absence in this part is undoubtedly connected with the slight development of the furrow.

Figure 22.

In the series of segments shown here there are a number of more or less incomplete furrows, and each is accompanied by abnormalities in the genital organs. The figure is a view from the ventral side.

The two furrows between the segments *a* and *b* correspond closely in position, except at the right, where the ventral furrow turns forward just before reaching the edge and ends on the ventral surface, but the dorsal furrow continues to the edge, where it ends. At the left neither of these partial furrows reaches the edge, though both end near it, the dorsal furrow nearer than the ventral. The genital organs at the right of these two segments, *a* and *b*, appear normal in form and position. The ovaries are the normal distance apart, as might be expected from the character of the furrows in the ovarian region. The two pores are equidistant from the dorsal furrow between them. At first glance the right pore in *a* appears to be quite close to the anterior boundary of the segment, but it should be noted

that the notch in the right edge which appears to separate *a* from *b* is really in *a*, for the dorsal furrow reaches the edge anterior to it, and the two pores are equidistant from this furrow. At the left side of *a b* there are two complete sets of organs opening to the exterior through a common pore which lies in the middle of the undivided edge of *a b*. In the region of the ovaries the furrows are normal, and accordingly the ovaries are nearly the normal distance apart. The left end of *a* is short, so that the full normal distance between the ovaries is not attained. Since the

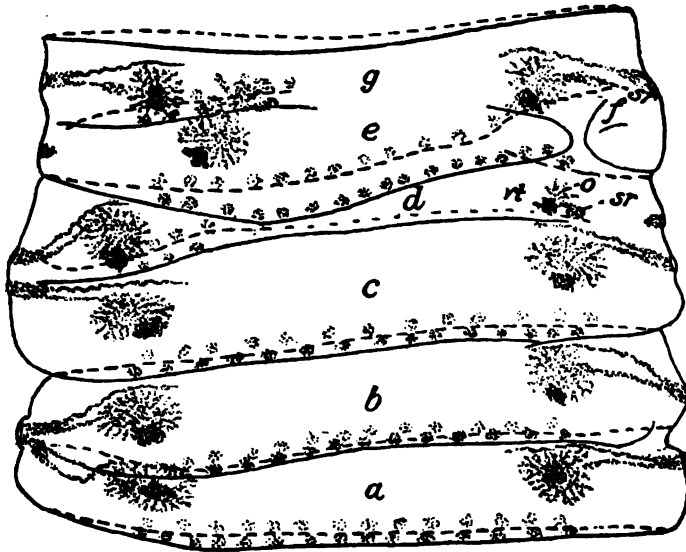


FIG. 22.

furrows do not quite reach the edge, it is undivided, and presents the relations of a single rather long proglottid. As might be expected, only one pore is present, though rather larger than normal, and into this the two oviducts and two vasa deferentia open, for the two sets of ducts approach each other as they reach the undivided region.

The segment *c*, of about normal length, and the segment *d*, which is of less than the normal length, except at the edge, are partially separated by two partial furrows which correspond very closely in position and extent on the two surfaces. The segment *c* is nearly the same length throughout, but *d* is longer at the

left edge than at the right and is very short in the middle region, and especially just to the left of it, in consequence of the bent course of the furrows bounding it anteriorly. At the right the two furrows between *c* and *d* end on the surface in the region of the ovaries, so that the right edge is undivided. At the left the furrows end very near to the edge, but do not reach it. The organs of the right side in *c* are normal; in *d*, however, they are rudimentary, consisting of a small imperfect ovary, *o.*, vitellarium, *vt.*, and a small closed and empty seminal receptacle, *s.r.*, and, entirely unconnected with these, a pore at the edge. No male organs except the cirrus appear. The absence of male ducts is perhaps due to the fact that this region of the segment *d* is shorter dorsally than ventrally, in consequence of the peculiar arrangement of furrows anterior to it. The pore of *d* is approximated to the pore in *c*, apparently because the furrows do not reach the edge. At the left edge *d* is of nearly normal width, though it narrows rapidly from the edge inward. Corresponding to its size the set of organs is of about normal size, like the left organs in *c*. These two sets open by distinct pores, which are, however, approximated.

The furrows anterior to *d* are very irregular. From the left edge they extend slightly posteriorly, thus almost separating *d* into two parts, then bend forward again and on the right end in a peculiar manner. The ventral furrow does not reach the right edge, but bends anteriorly and back upon itself, and ends on the surface. From the right edge the other portion of the furrow extends inward for a short distance, then curves back upon itself and ends. In the concavity of the curve near *f* a short isolated furrow appears. The furrow on the dorsal surface bends further anteriorly as it approaches the right edge and finally ends on the surface before reaching it. Posterior to this furrow lies another partial furrow corresponding to the right portion of the ventral furrow. It does not, however, bend back upon itself, but extends some distance to the left and then ends free on the surface. Thus a small region, *f*, is incompletely marked off as a partial segment on the dorsal surface, but ventrally the curved furrows divide into two parts. No genital organs appear in *f*.

The regions *e* and *g* are partially separated at the left by cor-

responding partial furrows, but at the right there is no separation, unless the region *f* be regarded as representing the right side of *e*. It is perhaps more correct to say that the right side of *e* is bounded ventrally by the left one of the two curved furrows, while dorsally *e* runs into *g*, and a small partial segment, *f*, mostly dorsal, laps over the edge to the ventral surface and fills the space left.

The furrows separating *e* and *g* on the left half of the body do not reach the left edge, though they end very near it. They are both rather shallow, thus indicating that the division between the two partial segments is less complete than normal.

The genital organs at the left of *e* present very peculiar and unusual relations. Ovary, vitellarium, and seminal receptacle of the normal size are present, and anterior to these and extending into *g* is a vas deferens which is closely coiled. This whole complex of organs does not lie in the normal position, but somewhat to the left of it, in the longest region of the partial segment *e*. I think it is possible that its position is due to the fact that the length is greater here than elsewhere. There is no trace of ducts leading to the edge; indeed, the oviduct beyond the seminal receptacle is absent, and the vas deferens does not extend even beyond the edge, but is coiled in a mass just anterior to the ovary. Doubtless this condition is due to the abnormally great distance between the organs and the edge. Careful examination of both surfaces of the region about the organs showed that there was no trace of a surface pore. I found, however, that the seminal receptacle was full of spermatozoa, a fact which indicates that the vas deferens opens directly into the seminal receptacle. The coils of the vas deferens were so dense and close, however, that it was impossible to find the connection. A pore corresponding to this set of organs exists at the left edge of *e*, but it is rather small and there is no trace of ducts leading from it. At the left of *g* there is a normal set of genital organs. On the right side of *e g* there is no division, unless, as suggested above, *f* be regarded as corresponding to *e*, and accordingly only one set of genital organs appears. In this the vas deferens is complete and normal, but the oviduct does not connect with the pore at all, ending instead with the seminal receptacle, *s.r.*, which is of

, nearly normal size, but empty. It will be noted that furrows bounding *f* anteriorly end very near the region of the oviduct on both surfaces, and it seems probable that this abnormal condition is due to the position and direction of these furrows. The position of this set of organs as a whole is normal, and since the anterior of the two dorsal furrows does not reach the edge we find the pore in the middle of the edge *f g*.

The region *f*, although as large as some partial segments which possess at least rudimentary genital organs, shows no trace of any such. The ventral surface is cut by furrows in various directions, and this is probably the reason why no ovaries appear. The single pore at the edge of *g* is just between *f* and *g*, for the two are not separated at the edge, so that there was probably no tendency for another pore to arise in *f*.

Inter-proglottidal glands appear in all the transverse furrows which lie within the region of their occurrence, except the furrows between *c* and *d*. Here the glands appear only near the left ends of the furrows, but these portions are deeper and thus more nearly normal than the rest of these furrows.

Figure 23.

This figure, a dorsal view, shows two segments at a later stage of development, in which the uterus is formed, and the other

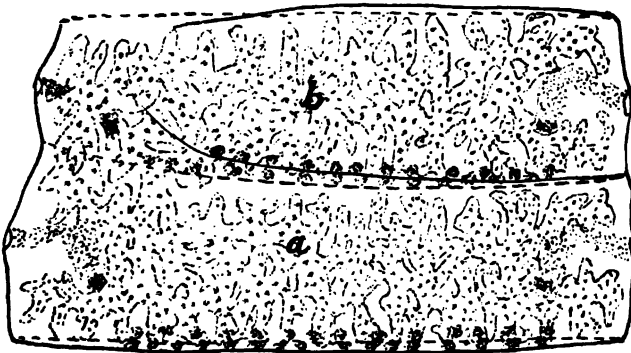


FIG. 23.

genital organs have undergone degeneration. The two segments are incompletely separated at the left, the ventral furrow being

shallow, but reaching the edge, and the dorsal furrow curving anteriorly and ending on the surface. The uterus is drawn in this case, and it is seen that the uteri of the two segments are continuous at the left, where the separation of the segments is incomplete. This is the only case of this kind figured, but continuity of the uterus is common in cases of partial division.

Regarding the other genital organs little can be said, as they are far advanced in degeneration. All appear to have been normal except at the left of *b*, where the pore and vitellarium are still visible, but no traces of ducts appear, and only a few cells in the ovarian region. A more or less rudimentary condition of these organs might be expected, for this portion of the segment is of less than normal length and dorsally is not marked off from either of the segments adjoining.

HULL ZOOLOGICAL LABORATORY,
UNIVERSITY OF CHICAGO,
April, 1900.

A DESCRIPTION OF THE MALE OF PERIPATUS EISENII WHEELER.¹

AUGUSTA RUCKER.

THIS new *Peripatus* from Tepic, Mexico, was named by Dr. W. M. Wheeler, of the University of Texas, and the female was described by him in the *Journal of Morphology* for October, 1898. It is from his material, which Dr. Wheeler has placed in my hands, and under his guidance, that I have obtained the following results. In this paper I have undertaken to give a description of the general external character of the male as differing from the female, and a description of its reproductive organs, with a brief account of the spermatophores. I hope in a short time to follow this up with the anatomical details, and later on to give the embryology of this most interesting animal.

The males proved to be so abundant in the material that an excellent opportunity presented itself for the study of this sex. Out of the original number, consisting of eighty-six specimens, thirty-two, on close examination, were found to be males. They are very much smaller than the mature females; in fact, several of the mature males in the material were smaller than embryo females (2 cm. in length) which I removed from the uterus. The largest of the males measured only 2.8 cm., while the largest female was 5.8 cm. in length. As to whether the sexes differ in color I cannot say, since a glycerine preparation used in softening the animals for sectioning has removed much of the color.

It has already been shown that this species varies in the number of its walking appendages, as do all the other well-known neotropical forms. The highest number of legs for *Peripatus Eisenii* is twenty-nine pairs, while the lowest is

¹ *Contributions from the Zoölogical Laboratory of the University of Texas*, No. 5. Director W. M. Wheeler.

twenty-four pairs.¹ Sedgwick's statement that the males have the lowest number of legs holds good here in every case except one. All those specimens having twenty-nine, twenty-eight, or twenty-seven pairs of legs were females, while all those with twenty-six, twenty-five (with one exception), or twenty-four pairs were males.

The number of these appendages is fixed at birth, as is also the case with *P. Edwardsii*, as Sedgwick has shown, and the

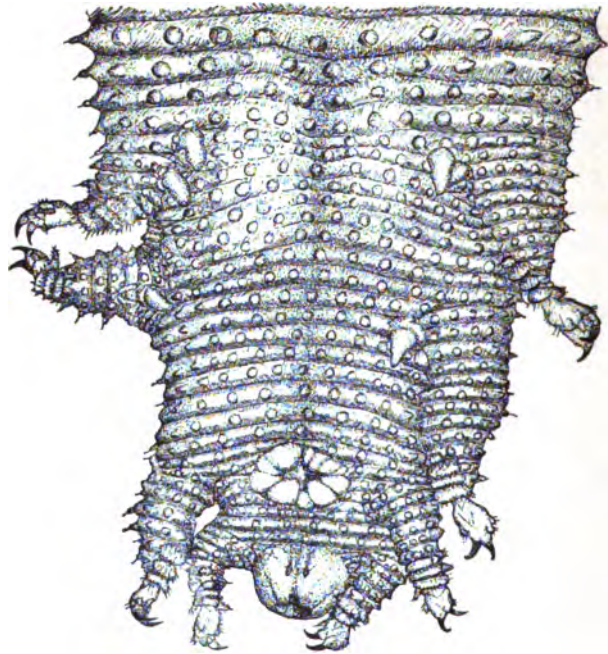


FIG. 1.

number of appendages of the mother is not necessarily transmitted to the embryo. From a mother with twenty-seven pairs of legs, three embryos were removed, each with twenty-eight pairs of appendages; and again from a mother with twenty-eight pairs of legs three embryos were removed, the two most mature of which had twenty-five pairs, while the less mature one had twenty-six pairs. These last three specimens had

¹ Dr. Wheeler mentioned in his paper one specimen with twenty-three pairs of appendages; this I was unable to find after carefully reexamining all the males.

other external characters, apart from the number of appendages, in the appearance of the posterior portion of the body, which, embryos as they were, showed them to be males.

The anterior legs of the males are like those of the females, each possessing four pads and a pedal groove. The nephridial opening is on the second pad from the base, on the fourth and fifth

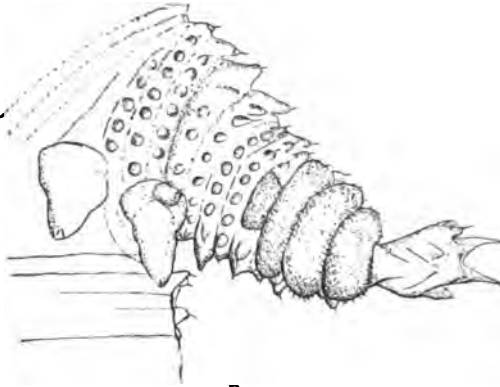


FIG. 2.

leg, as in the female. On the third posterior leg the proximal pad becomes much reduced and entirely disappears on the penultimate leg, while on the last leg only the two distal pads remain, with a portion of the original second proximal pad.

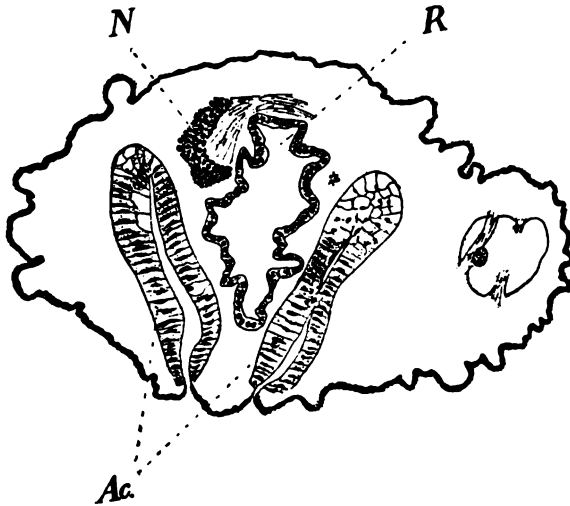


FIG. 3.

On the four posterior pairs of appendages there are no pedal grooves, and here begins the difference between the appendages of the two sexes. In the place of the pedal grooves, which are entirely wanting on the third and fourth posterior

legs, there are two long, soft papillae for each appendage. This is invariably the sign of the male *Peripatus Eisenii*, and these papillae, with an opening at the tip for the outlet of the crural

glands, are always on the third or fourth posterior appendages. The only specimen with twenty-five pairs of legs, which did not have these tubercles, was opened and found to be a female. The position of these papillae can best be seen from Fig. 2, which is a camera lucida drawing of the left fourth leg. Fig. 1 is a drawing of the ventral surface of the posterior end of an animal 2.4 cm. in length. At first sight the fourth or third leg may appear to have only one papilla or none at all, but on closer examination the tip of the papilla will be seen to be surrounded by a circular ridge. Sections through these posterior legs of different individuals show that the tubercles are of uniform development in all males, and that they can be retracted or protruded in the living animal. The section also shows that the papillae are retracted only by involuntary muscle fibers inserted on these papillae. Fig. 4 represents a section through the fourth leg. The inner papilla is protruded, while the outer

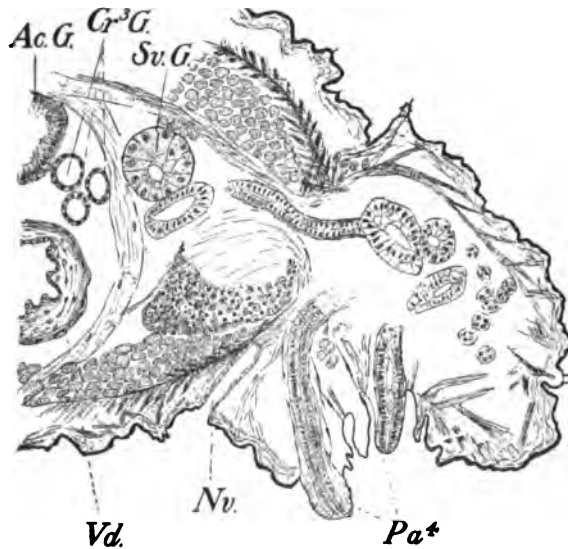


FIG. 4.

one is partially withdrawn. Fig. 5 is a section through the third leg, showing the inner tubercle more retracted than the outer one in Fig. 4. This tubercle has about reached its limit of retraction. The outer papilla was cut to one side, so as to

show the cup-shaped depression in which it rests and the distinct outline of the epidermis which makes it look like a diminutive pine cone.

The opening of the generative organs is, like that in the female, between the penultimate pair of legs; but this pair of



FIG. 5.

appendages, unlike that of the female, has no trace of a pedal groove, and the same may be said of the last pair. There is likewise no trace of the nephridium in the penultimate pair of legs, whereas the last pair possesses these organs, which appear in section with small external openings.

Just as there are crural glands in the male, which are wanting in the female, so also are there *accessory glands*. There is a pair of these glands which opens externally by two small slits situated between the generative and anal openings, about a fourth of the distance from the latter. Fig. 3 represents a camera drawing of a section through the orifices of these accessory cells.

Before leaving the consideration of the exterior of *P. Eisenii* it is well to speak of a thing of interest which relates to both the sexes, and which has not been mentioned before in connec-

tion with this species. It is a bean-shaped papilla that is always found in a depression on the dorsal surface of the leg where it joins the foot. Gaffron describes this papilla in *P. Edwardsii*. Sedgwick says it is also found in the Trinidad species and is probably characteristic of all the neotropical forms. The surface of the depression in which the papilla lies is smooth, while the papilla itself shows a distinct cell structure, the cells all converging



FIG. 6.

toward the center. Fig. 6 represents a longitudinal section through the foot splitting the papilla.¹

From the number of external outlets of glands connected with the generative tract, it is readily seen that the male repro-

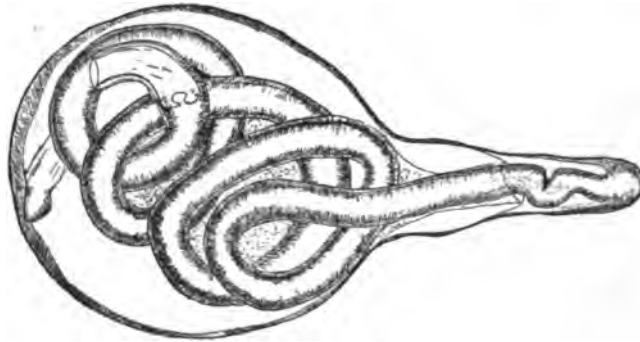


FIG. 7 a.

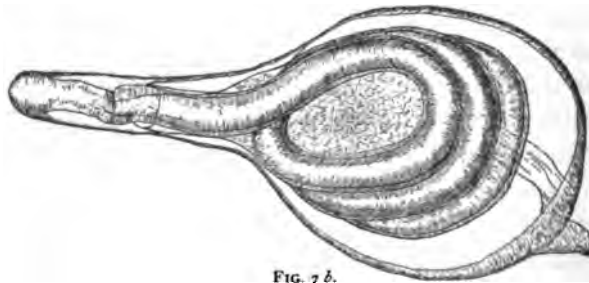


FIG. 7 b.

¹ It seems from the position of these papillae, especially when the foot is drawn in, that they are sensory. If this be true, the comparison of the foot of *Peripatus Eisenii* with the parapodium of the Chaetopoda is rather striking, the sensory papillae corresponding to the cirri.

ductive organs are much more complicated than those of the female. The latter has two fused ovaries, paired receptacula ovarum, paired receptacula semines, and paired uteri. The testes are large tubular organs beginning at about the posterior third of the body and running backward without much twisting to the seminal vesicles, which are somewhat larger in diameter. The seminal vesicles appear as dilatations of the testes, the right one of which is some distance in front of the left. The vesicles of all the specimens I have examined are full of the spermatogonia discharged from the testes, spermatocytes, and spermatozoa. The material was collected in October, when the testes were active. The seminal vesicles lead posteriorly into a

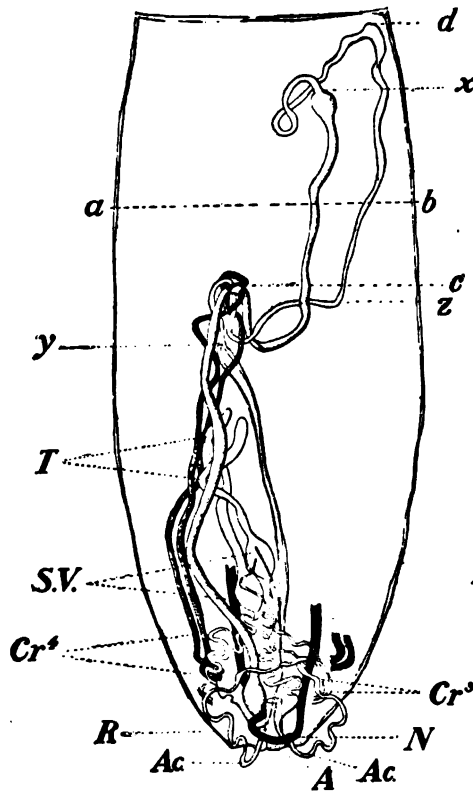


FIG. 8.

pair of exceedingly convoluted vasa deferentia, the right one of which passes over and then under the nerve to join the left, which passes under the nerve. They then run forward side by side, as two small, very thin-walled, straight ducts, for some distance, till they unite to form a common duct. These paired portions of the vasa deferentia are quite full of spermatozoa. The unpaired portion of the testicular ducts is of great length, sometimes exceeding twice the length of the whole body. This tube is clearly divided into two portions, the first

two-thirds of which are comparatively thin walled and lined with ciliated cells, while the last third has a non-ciliated epithelial lining and very thick muscular wall. This thick-walled portion terminates in an enlarged sack which might well be called the spermatophore sack, since it holds a spermatophore in nearly all the specimens examined. The sack opens on the exterior by means of the generative orifice between the

penultimate pair of legs.

The portion of the vas deferens possessing the thick muscular wall does not constitute the spermatophore maker, as Moseley found in *P. N. Zealandiae*, but it is the thin-walled portion which has this

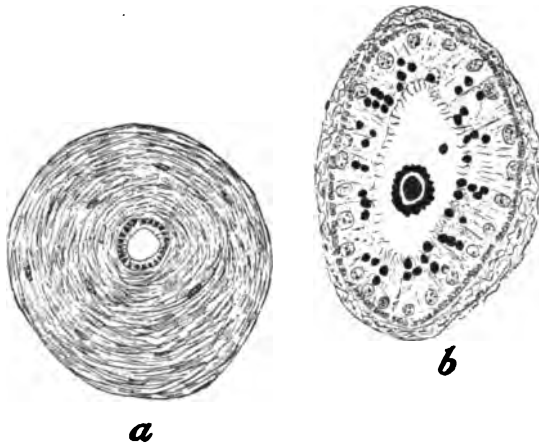


FIG. 9.

function, though the epithelium is ciliated in that region.

Fig. 8 is a partially diagrammatic drawing of the male reproductive organs of *Peripatus Eisenii*. The vas deferens from *c* to its termination has a thick muscular wall. From *y* to *d* and thence back to *c* the wall is comparatively thin, and the lining cells are at the same time ciliated and secretory. Especially active are the cells of that portion which begins at about *z* and ends at *d*. Here the most substantial portion of the spermatophore is made. In secretion great balls of a glutinous substance staining dark in haematoxylin are formed in the inner cells and given off. They become packed into the spermatophore rod in the most regular manner, making its surface appear to be marked off in regular hexagons, not indicated in my drawing. This rod receives lighter secretions as it passes along, carrying before it a packet of spermatozoa around which it becomes very much coiled in the dilated distal portion

of the vas deferens, or spermatophore sack. Here the coiled spermatophore seems to receive other layers of secretions which form a case of some thickness. Fig. 7 and 7 a are camera drawings of two views of a spermatophore, the pointed end of which projects forward in the vas deferens.

The crural glands which open out through the above-described papillae are found only in the male. These glands from the fourth pair of legs are large and extend almost half the length of the animal. They leave the lateral compartment of the body (unlike the same glands of *P. capensis*, which run their whole distance in this portion of the body) almost immediately to coil around the vas deferens. The crural glands of the third pair of legs are very thin tubes winding in and around the convoluted portions of the vasa deferentia, and around the seminal vesicles, where they end. The accessory glands are large tubes which are situated dorsally to the other organs; they run posteriorly (the right one going over and just under the nerve), to empty a very short distance in front of the anus.

In concluding this description, one point of great interest presents itself which cannot be overlooked. *This is the rapid sexual development of the males to maturity.* I observed that in sections of very small specimens which could not have been long from the uterus, the seminal vesicles were distended with ripe and rapidly developing spermatozoa. In a male embryo which was removed and sectioned, I found in the seminal vesicle not a few spermatozoa and spermatids in abundance. It would seem to follow from these conditions that the males of the neotropical species of *Peripatus* must be rather short-lived, and this fact will probably account for their scarcity.

UNIVERSITY OF TEXAS, AUSTIN, TEX.,
May 1, 1900.

BIOLOGICAL BULLETIN.

ABNORMALITIES IN THE CESTODE MONIEZIA EXPANSA. II.

C. M. CHILD.

I. *Spiral Abnormalities.*

IN the cases described in this section spiral modifications of the segmentation are present in greater or less degree. Associated with these are often found examples of partial division resembling those described in Part I, *Biological Bulletin*, Vol. I, No. 5. Where these are closely connected with the spirals they are shown in the figures and briefly described.

Figs. 24, 26, 27, 30, 38, 39, are selected from a number of different individuals. The other figures are all taken from the single worm mentioned in Part I as possessing a very large number of abnormalities. Figs. 34 and 35, being taken from a point nearer the anterior end of the chain, where the size is much less than in older proglottids, are magnified about fifty diameters, the other figures about twenty.

For terms used in the description, the structure of the normal segment, etc., the reader is referred to the first paper (*Biol. Bull.*, Vol. I, No. 5).

Figure 24.

The principal feature of this figure is a case of partial division, which is in reality a short spiral. The proglottid *a* shows at the right a very short furrow extending from the edge a short distance over the upper surface and ending free.

The lower surface shows no corresponding furrow. The length of the proglottid at this side is somewhat greater than, but not double, the normal length, *i.e.*, it is not as long as two fused proglottids of the same age. Two groups of cells, the "Anlagen," of the reproductive organs or "genital masses," appear upon this side, however, as would be the case if the short partial furrow were complete. The furrow itself indicates the imperfectly double character of the segment, and the two genital masses show this still more clearly. At the left *a* is only half as long as at the right and possesses only a single genital mass. The partial segment *b* is completely separated from *a* both on the upper and lower surface, but is seen to be

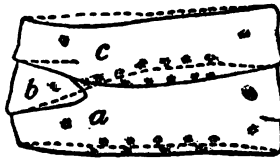


FIG. 24.

connected with *c* on the lower surface. The furrow separating *b* from *a* runs inward and somewhat anteriorly from the left edge for about one-third the width of the body, then turns and extends outward and anteriorly until it joins the complete furrow in front.

Thus the small piece *b* is completely marked off on the upper surface, and though its edge at the left side is of normal length, it narrows to a rounded end. On the lower surface the relations are different, for the partial furrow between *b* and *c* on this surface ends free, while the complete furrow separating *a* and *c* at the right bends so as to pass posteriorly to *b* at the left and connects at the left edge with the furrow between *a* and *b*. The partial segment *b* is thus a short spiral, making less than half a turn. Notwithstanding its small size, it shows a genital mass as large and distinct as any at this stage.

Figure 25.

Here two examples of partial division and a short spiral occur. Upon the upper surface the two partial proglottids *a* and *b* are incompletely separated, the partial furrow on the left side being longer than that on the right. The partial furrows at the right correspond exactly on the two surfaces, both ending free. The partial furrow on the upper surface at the left forms

the beginning of a spiral furrow which makes one and a half turns. It is oblique upon the lower side, running from between *a* and *b* at the left to the anterior edge of *b* at the right, then passing over the upper surface again as a complete transverse furrow anterior to *b*, and finally ending free on the lower surface. Thus the spiral segment *b* is open at both ends. If the furrow between *a* and *b* on the upper surface were complete, the spiral would begin between *a* and *b* at the right on the lower surface, and the furrow would thus make almost two turns. In the region where the genital masses appear the furrows show very nearly normal relations, and the position of the genital masses needs no comment.

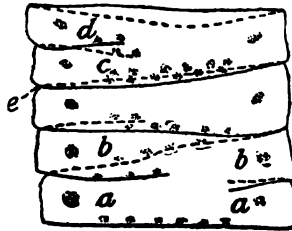


FIG. 25.

In *c d* another case of simple partial division occurs at the left, the partial furrow corresponding in position on the two surfaces. At the left two genital masses occur, while at the right, where *c d* is undivided and shorter than at the left, only one appears. All the partial furrows which extend far enough from the edges to lie within the region where the interproglottidal glands occur, possess them. The furrows between *a* and *b* on the right show none, as they are too short.

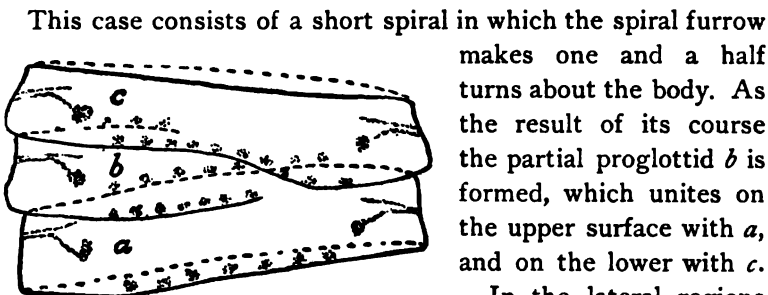
Figure 26.

FIG. 26.

This case consists of a short spiral in which the spiral furrow makes one and a half turns about the body. As the result of its course the partial proglottid *b* is formed, which unites on the upper surface with *a*, and on the lower with *c*. In the lateral regions the segmental boundaries are all normal, and, accordingly, the organs are situated nor-

mally, but at the left there are three segments and at the right only two, and a corresponding number of sets of genital organs is found.

Figure 27.

A spiral furrow making only a little more than half a turn appears in this case. At the left the upper surface of *a* is united at the edge with the lower surface of *b*, and at the right the lower surface of *a* unites at the edge with the upper surface of *b*.

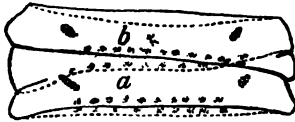


FIG. 27.

The only abnormality visible at this stage in the genital organs appears at the left in *a*. Here the genital "Anlage" is elongated and narrower than in the other cases. The proglottid is not sufficiently developed to show the ducts and pores, so that it is impossible to determine just what the situation of these organs will be.

Figure 28.

Here the natural relation of the dorsal and ventral surface is somewhat altered. The figure is drawn with the dorsal surface uppermost, and it is seen that the furrows on the dorsal surface lie further posteriorly than those corresponding to them on the ventral surface. The furrows bounding *a* posteriorly do not meet at the edge, as they would if normal and merely distorted by pressure or otherwise, but the end of the ventral furrow is anterior to the dorsal. The furrows *d* and *d'* would correspond to each other if normal, but as a matter of fact *d'* meets *e* at the left edge instead of meeting with its corresponding furrow *d*, thus producing a slight spiral. The furrows *e* and *e'* would meet at the two edges if normal, but here again the ventral furrow is considerably anterior to the dorsal except at the right edge, and its left end shows no indication of bending posteriorly to meet the latter. A somewhat similar condition is seen frequently in mounted specimens, but in most cases is simply a distortion due to the compression between glass plates during fixation. The real abnormalities such as occur

here can be distinguished by the fact that at one edge or the other or both the corresponding furrows on the two surfaces do not meet. That some distortion has also occurred in this case is probable from the fact that in the regions immediately outside and posterior to that of the figure otherwise normal segments are oblique dorso-ventrally, as if the dorsal surface had moved posteriorly over the ventral, or the ventral anteriorly over the dorsal. The segment *b* is bounded by a furrow beginning at *d''* and forming a spiral of nearly two turns, ending free on the ventral surface (*e'*). The genital organs *b'*, on the left side of *b*, lie almost between the dorsal furrow *e* and the ventral *d'*, and it is the only genital mass on the left for the whole spiral. The ovary and vitellarium, being nearer the ventral surface, appear between the furrows *d'* and *e*, while the ducts lying nearer the dorsal surface bend posteriorly, and their terminal portions appear posterior to the furrow *e* on the dorsal surface, and finally reach the surface almost midway between the dorsal furrows bounding *b*. At first glance it ap-

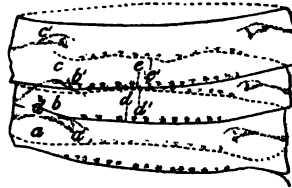


FIG. 28.

pears that if the position of the genital organs is correlated with the form of the proglottid, the duct should open somewhere in the region *c* instead of passing posteriorly under the dorsal furrow *e*, as it does. As a matter of fact, however, its position is the only one possible in the spiral proglottid *b*. Since the spiral segment *b* lies somewhat obliquely, *i.e.*, with its ventral surface somewhat anterior to the dorsal, the position of the organs at *b'* between the dorsal furrow *e* and the ventral *d'* is only apparent. In reality they are in about the normal position in their segment *b*. The outer end of the ducts is rudimentary, consisting of a scarcely visible strand of cells, and there is no enlargement in the region of the pore. Moreover, the inner end of the vas deferens instead of running anteriorly to the ovary and vitellarium, as is usual, is posterior to them, as seen in the figure (*b'*). This position of the inner end of the vas deferens posterior to the ovary is peculiar and is probably due to the oblique position of the segment *b*.

The rudimentary character of the terminal portion of the ducts is apparently due to the fact that the proglottid *b* is not wholly distinct from *a* in this region. The posterior dorsal furrow is interrupted at *d''*. The short portion extending to the left edge is not a furrow of normal depth but a scarcely visible fold upon the surface, and the left end of the main furrow at *d''* is also very shallow. On the ventral side there is no furrow corresponding exactly to the furrow *d'd''*, for *d'* is a spiral continuation of it. Therefore the only evidences of separation between *a* and *b* in this region are the slight furrows at *d''*. Since the degree of separation is so slight, the tendency to form a second genital pore and the terminal region of the ducts is probably very slight also, but is still present, as is evident from the figure. The testes are just beginning to appear (not represented in the figure), and their distribution corresponds exactly with the conditions on the dorsal surface.

Figure 29.

At the stage shown here the proliferating groups of cells forming the reproductive organs are visible, and the interproglottidal glands are more numerous. The variation is a spiral, the furrow making two complete turns. The posterior end of the spiral furrow appears between

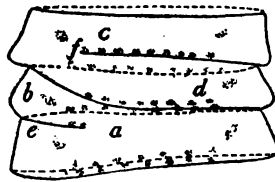


FIG. 29.

a and *b* at *e*. Upon the lower surface it is a complete furrow and is continuous with the furrow upon the upper surface between *b* and *d*. This bends anteriorly at the left instead of completing the furrow between *b* and *a*, and so separates *b* and *c* at the left edge, continuing over the lower surface as a complete furrow and passing once more to the upper surface between *d* and *c* at the right and finally ending free at *f*. The only portion of this continuous furrow which differs greatly in position from the normal is the part between *b* and *d* at the left on the upper surface, where it bends anteriorly and so fails to complete the separation between *a* and *b*. The spiral is the result of this bend

in the furrow, but since the furrow ends free at *e* and *f* both ends of the spiral proglottid bounded by it are open and connect respectively with *a* and *c*. The position of the genital masses is not affected by the presence of the spiral arrangement.

The general relation of the inter-proglottidal glands to the furrows is shown by the fact that the glands appear with the abnormal and partial furrows as well as with the normal.

Figure 30.

This figure shows two spirals situated in the regions designated by *a* and *b*. In each case the spiral is due to the curving of the furrow near the median line of the body. In the one case the curve is on the dorsal surface, in the other on the ventral. Since the curved furrows are on opposite surfaces and yet nearly parallel, the two spirals are opposite in direction. In both cases the ends of the spiral portions unite more or less completely with adjacent segments, owing to the fact that the furrows between them end free.

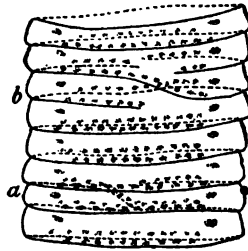


FIG. 30.

In *b* this union is much more complete than in *a*. In *a* the spiral furrow makes two turns about the body, in *b* only one and a half as a continuous furrow. If, however, the partial furrows at the right in *b* be considered as a continuation of it, this furrow also makes two complete turns.

The lateral regions and edges of the segments are all normal in form, and we find all the genital masses normal in position.

Figure 31.

The figure is a view from the dorsal side of a series of segments, showing a number of abnormalities. The first of these is the small partial segment *b*, wedged in at the right between *a* and *c*. Its dorsal surface is greater than its ventral, and its edge is nearly as long as that of a normal segment. Dorsally the furrow between *a* and *b* ends free on the surface. The

corresponding ventral furrow turns anteriorly a short distance from the edge and meets the main furrow between *a* and *c*. Thus the ventral surface of *b* is completely marked off from other segments. Both the dorsal and ventral furrows between *b* and *a* are rather shallow. The organs in *b* are distinctly abnormal. A rather small ovary and vitellarium appear nearer the edge than in normal segments, probably because of the increasing length of the segment *b* nearer the edge. The oviduct is incomplete and ends bluntly, as the figure indicates.

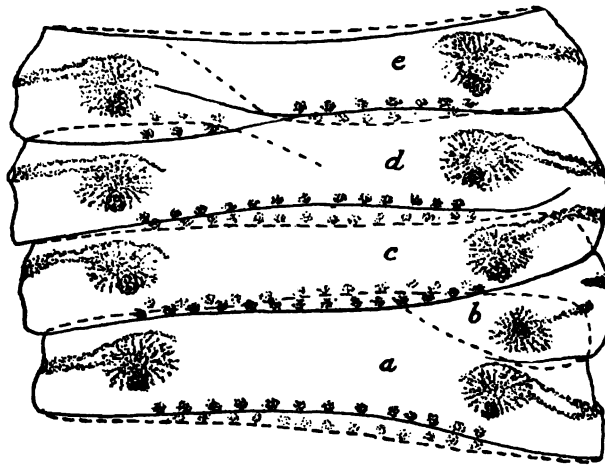


FIG. 31.

A distinct pore is present, and connected with it is a well-developed cirrus, but no trace of a vas deferens is found anywhere in the segment. The position of the various organs illustrates well the relation of each to the form of the segment in the region where it occurs. Thus the ovary and vitellarium, which appear from the dorsal surface to be near the posterior edge of the segment, lie midway between the bounding furrows on the ventral surface, and the oviduct, which extends somewhat dorsally from the ovary, runs obliquely forward towards the edge, so that it is normally placed with regard to the furrows on the dorsal surface. The pore lies near the anterior end of the segment. The ventral furrow bounding *b* anteriorly turns backward before reaching the edge and unites with the

posterior furrow, so that the region corresponding to the ventral side of *b* is cut into two parts, that nearest the edge being united with *c*, *i.e.*, taking *b* as it appears on the dorsal surface, we find that it is not separated from *c* at the edge. As *b* is bounded on the ventral surface it does not reach the edge of the body at all. The position of the pore is evidently connected with these peculiar relations. The dorsal side of *b*, together with *c*, forms a spiral. Beginning with the dorsal partial furrow between *b* and *a*, the spiral furrow makes two complete turns about the body.

The rudimentary condition of the organs in *b* is undoubtedly due to the small size of the segment. The ovary and the oviduct are more completely developed than the vas deferens. The segment *b* contains a number of testes and in some spermatozoa are visible.

The position of the organs in *a* needs no comment. The segment is of peculiar form, owing to the presence of *b*, but its genital organs are normally situated.

The furrows between *c* and *d* are abnormal. The dorsal furrow ends at the right without reaching the edge, and the ventral furrow turns posteriorly near the right edge and meets the posterior boundary of *c*. Thus the right edge of *c* and *d* is not divided by any furrow, but the dorsal furrow extends almost to the edge. The ovary and vitellarium at the right of *c* are normally placed with regard to the ventral boundaries, and the ducts and pore with regard to the dorsal boundaries. Both ducts cross the course of the ventral furrow at an angle to reach the edge, thus indicating that relations on the ventral side have little influence on their direction. At the right of *d* a normal set of organs occurs. The figure shows, however, that the two pores on the right edge of *c d* are near together. In the region of the inner ends of the ducts the segments are completely separated, and the distance between the two sets of ducts is normal here. As they approach the surface, however, the separation between the two segments on the dorsal surface becomes less and less complete, and the edge itself is undivided. Thus the pores tend to form near its middle, but the fact that the dorsal furrow extends so nearly to the edge

indicates that a certain degree of individuality exists up to and perhaps beyond the point where it terminates, and this, together with the length of the edge of *c d*, accounts for the presence of two pores instead of the union of both sets of ducts in a single pore.

Between the segments *d* and *e* the furrows are very abnormal. The ventral furrow is divided into two parts which overlap on the surface, the one turning anteriorly, the other posteriorly. The oblique portions are very shallow and do not bear interproglottidal glands.

The dorsal furrow is also in two parts. The one at the left does not turn posteriorly, but continues as a very shallow furrow over the region corresponding to that which the ventral furrows leave undivided, and finally unites with the right half. This latter, however, continues to the left, beyond this point, but turns anteriorly, running up into the segment and ending just dorsal to the ovary. The oblique portion is shallow, like the oblique portions of the ventral furrow, and bears no glands. The genital organs at the left of *d* and *e* are normal, however, doubtless because the growth has been normal in the regions where the organs occur. Only the oblique portion of the dorsal furrow approaches the ovary, but, as has been repeatedly shown, the position of the ovary is influenced only very slightly, if at all, by the form of the dorsal surface.

Figure 32.

At the stage of development shown in the figure the genital masses are becoming differentiated into the various organs. The female portion is mostly distinct from the male, and the strands of cells forming the ducts extend nearly or quite to the edge of the body, though the pores are not distinct as yet. The variation shown is a spiral in which the furrow makes two complete turns, the spiral segment bounded by it making one complete turn. The spiral begins on the right in the short furrow bounding *a* posteriorly and separating it completely from the proglottid behind; from this point it passes around the body, bending forward at the right side of the upper sur-

face to form the anterior boundary of *a*, then making one more complete turn and ending on the upper surface, thus leaving *b* and *c* incompletely separated at the right of the dorsal surface. The development of the genital organs is sufficiently advanced in this case to show the very intimate relation of these organs as regards position with the form and relations of the proglottids. The segment *a* possesses its own genital mass (*a'*), which is entirely separated from all the others. This is, however, of less than the normal size and does not reach the edge of the segment. It is divided into two parts in its inner portion, but the group of cells which would later form the ovary and vitellarium does not appear. In fact, the mass seems to consist largely, if not wholly, of portions of the two ducts. It will be remembered that the ducts lie farther dorsally than do the ovary and vitellarium. The figure is drawn with the dorsal surface uppermost, and it is only dorsally that the region *a* appears as a distinct partial proglottid. On the ventral surface the relations of the furrows are entirely different. It appears then that the dorsal re-

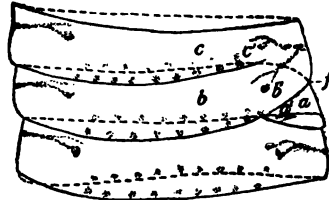


FIG. 32.

gion of *a* possesses a degree of individuality sufficient to cause the appearance of the organs proper to this region. The ventral region not being separated from *b*, the organs of the ventral side do not appear. Whether the organs would in later stages approach or reach the normal development it is impossible to state with certainty, but the evidence seems to be against such a view, for in all cases of similar abnormalities in much later stages the genital organs or parts, however rudimentary they may be, show the same degree of differentiation as those of normal segments.

In the large, incompletely separated segments *b* and *c*, there appears another example of the close relation between the individuality of the segment and the presence and arrangement of the reproductive organs. At the left appear normal sets of organs in normal position. At the right, however, where the furrow on the dorsal surface is incomplete and that upon the

ventral surface bends posteriorly, two sets of organs (b' and c') appear whose ducts open into a common genital pore. Each of the sets is apparently complete, possessing the groups of cells which will form ovary and vitellarium, as well as the vas deferens. The inner portions of these two sets are situated much as they would be if b and c were normally separated from each other, *i.e.*, their position is nearly normal. The partial furrow on the dorsal surface, however, does not extend to the right edge of the body, but ends free before reaching it, so that b and c are united here, and correspondingly only one genital pore appears at d , and into this both sets of ducts open. But the question now arises as to the reason for the connection of the organs b' with this pore. Normally these organs would open on the edge at some point not far from f , but, owing to the arrangement of the proglottids in this case, f is the point of intersection of the furrows, *i.e.*, does not possess the features of the region where the genital pore normally appears, for this is upon the edge, about midway between two furrows. The only possible conclusion from the facts is that the direction of the ducts and their final connection with the pore are correlated with the form of the proglottids in this region and especially upon the dorsal side. This conclusion is confirmed by the fact that the ducts cross almost at right angles a furrow on the ventral side, thus rendering it evident that their arrangement is not affected by its presence. In short, ovaries and vitellaria arise separately in b' and c' , because the relations of the ventral sides of the segments in that region are practically those which exist in two separate proglottids, and upon the dorsal surface the same is true in the immediate region of the inner portions of the organs. Nearer the edge, however, the relations on the dorsal side are those of a single segment, so that the two sets of organs approach each other and finally open in a common pore, which occupies a normal position with respect to the boundaries of the proglottid in its immediate vicinity.

Figure 33.

This figure shows a rather long spiral, together with a small completely separated partial proglottid. The furrows bounding the spiral begin between the partially separated segments *a* and *b* near the left side of the dorsal surface — the dorsal surface is uppermost in the figure — and make a little over three turns about the body. At the left side of the dorsal surface, between the segments *d* and *e*, the furrow becomes shallower, and on the left edge it terminates. The spiral segment enclosed by it makes a little more than two complete turns. In consequence of the course of the furrow, *a* and *b* are incompletely separated on the dorsal surface, but completely separated ventrally; the furrows bounding the regions *b*, *c*, and *d* do not correspond on the two surfaces, and finally *d* and *e*, which are distinct dorsally, are completely united on the ventral surface. These abnormal relations are accompanied by a number of corresponding abnormalities in the genital organs. At the left side *a* is distinct from *b*, and the genital organs *a'* on this side are normal and in normal position. At the left side of *b*, *c*, and *d*,

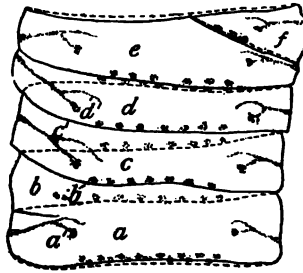


FIG. 33.

where the spiral character of these segments becomes evident, the genital organs show marked abnormalities. At *b'* only two small groups of cells are found, representing apparently portions of the ducts; at *c'* and *d'* full sets of organs occur, but lie obliquely, and the ducts are elongated. It is evident that the pores and the greater portions of the ducts are normal in position with regard to the dorsal form relations of the segments. The oblique direction of the ducts is apparently due to the fact that the dorsal side of *c* and *d* bends forward near the left edge. Since the inner portions of the organs are formed at the normal distance from the edge, in order to reach the edge as they do, the ducts must be longer than the normal, for they must run obliquely.

The dorsal sides of *b* and *c* both correspond in part, as

regards position, to the ventral side of *b*. The ovary of the set *c'* lies in *b* as bounded ventrally. Apparently the dorsal side of *c* and the ventral side of *b* are to be regarded as belonging together at the left, even though they do not occupy corresponding positions, as they do at the right. If this be the case, the organs *c'* show a close correspondence to the form relations. Upon the dorsal surface *b* is merely a small portion, intercalated, as it were, between *a* and *c* and incompletely separated from *a*. The genital organs are very rudimentary. The ventral organs found in *b* belong to the set *c'*, and there is no distinct ventral region corresponding to the dorsal side of *b*. Thus no ventral organs appear. Two small groups of cells (*b'*) are the only traces of genital organs in this region. These apparently represent portions of the ducts. This very slight development of genital organs is probably due to the small size and imperfect form of this portion.

The set of organs at *d'* shows much the same relations as that at *c'*. Its pore, however, is very close to the furrow between *d* and *e*, as is also the pore of the organs at the left of *e*, which are otherwise normal. The approximation of these pores is evidently correlated with the incomplete separation of *d* and *e* by a shallow furrow on the dorsal surface, and not at all ventrally.

On the right, at *f*, a small partial segment is separated from *e* by oblique furrows. It possesses a normal set of genital organs. The intercalation of *f* leaves the right edge of *e* very short, but the genital organs are, so far as appears, normal. Whether they will reach full development and normal size cannot of course be determined.

Figure 34.

The abnormalities figured here occur not far behind the scolex, where genital organs have not yet appeared. At *a* there is a small partial segment wedged in between two others at the left side. Just anterior to this is a spiral, beginning on the lower surface and making nearly two turns. The course of the spiral furrow is such that on the upper surface the segment *c* does not reach the left edge at



FIG. 34.

all, and on the lower surface the segment next to *a* narrows toward the left and ends at the edge.

Figure 35.

The figure shows a complex case of partial division (*a*) and a spiral of about four turns (*b c*). The region *a* is partially separated into two segments at the right, but at the left into three, the most anterior (*b*) forming the beginning of the spiral. On the lower side, just beneath *b*, there is a small region wholly marked off by furrows and not forming part of the spiral. The spiral *b c* is perfectly simple in form, though rather long. This case, like Fig. 34, was found near the anterior end of the chain, and neither genital organs nor inter-proglottidal glands are formed.

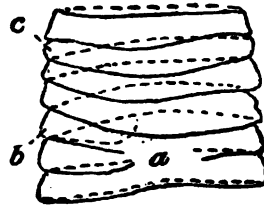


FIG. 35.

Figure 36.

The figure is a dorsal view of an extremely long spiral, which makes seven complete turns about the body. The spiral is due to the bending posteriorly of the ventral furrows near the right edge.

At the left the segments are all normal in form, and all of the genital organs are normally placed. At the right the curve in the ventral furrows produces complex relations in the various segments. All of the curved portions of the ventral furrows except the one anterior to *c* are much shallower than the transverse parts, as is indicated in the figure. In the one exception, the furrow anterior to *c*, the curved portion appears as distinct and deep as the rest of the furrow. In *a*, *b*, *d*, *e*, and *f* the inner portions of the organs of the right side are seen to lie in about their normal positions with respect to the boundaries of their segments. The ducts are parallel to the dorsal furrows and cross the course of the ventral furrows in each case, *i.e.*, they conform to the relations on the dorsal side. In the segment *c*, however, the ducts run nearly parallel to the

ventral furrows, crossing the dorsal furrow which forms the posterior boundary of *c*, and finally opening, together with the organs in *b*, into a single pore on the edge of *b*. This case appears to be an exception to the general rule of correlation between the arrangement of the genital organs and the form of the segment, for the ducts on the dorsal side cross the

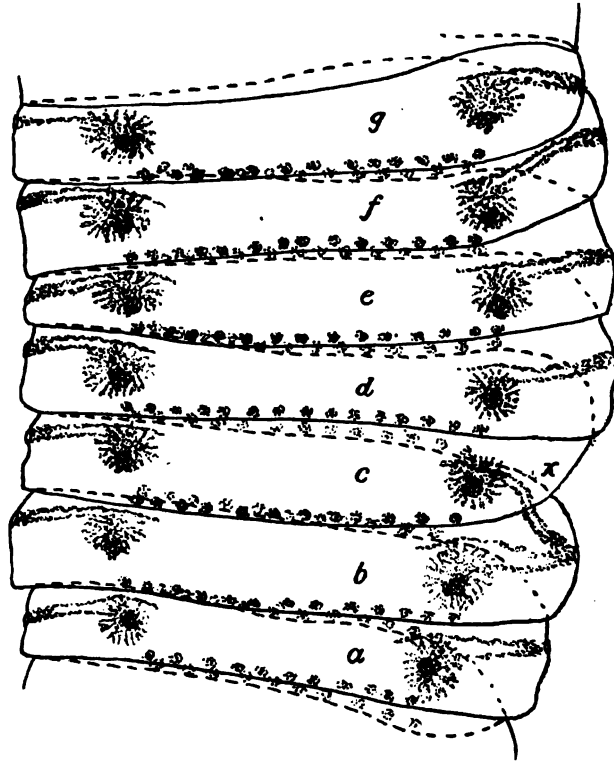


FIG. 36.

course of the dorsal furrow. As is evident from the figure, the ventral side of *c* and the dorsal side of *b* are very intimately connected at the right edge, more so than, for instance, the ventral side of *c* with the dorsal side of *d*. Moreover, the edge of *c* itself is oblique and very short, and the ventral furrow at *x* is deeper than the corresponding portions of the other ventral furrows. The course of the ducts from the organs at the right of *c*, differing as it does from the course of

the ducts in the other segments of the spiral, is undoubtedly determined by the relations existing here. Probably the small size of the dorsal side of *c* at the right is the real basis of the difference, for it is largely because of this that the ventral side of *c* is so intimately connected with *b* at the right.

The segment *g* is nearly normal in form in the region of the right ovary, and this lies in its normal position. Nearer the edge, however, the dorsal and ventral sides of the segment do not correspond, the ventral surface bending posteriorly, while the dorsal bends slightly in the opposite direction. The ducts and the pore evidently conform to the relations on the dorsal side, but they lie almost directly over one of the ventral furrows.

Figure 37.

This case comprises a number of segments which show an approach to the spiral form but do not quite attain it, since most of the furrows are not complete at the left. The figure is a dorsal view. It can easily be seen from the figure that if the furrows on the two surfaces were continued over the left edge, a spiral segment extending through the whole series would be formed. The manner in which a spiral arises is well illustrated by this case. The bending of the furrows near the edge on one surface is all that is necessary. Here the dorsal furrows bend anteriorly, while the ventral furrows remain straight, except between *a* and *b*, where there is a slight posterior curvature.

At the right the segments are all normally bounded, and the genital organs of the right side are normal in form and position. At the left, however, where the relations approach the spiral form, the organs show corresponding abnormal relations. At the left of *a* the anterior ventral furrow is normal in the ovarian region, but turns posteriorly near the edge, and the dorsal furrows bend anteriorly, so that the dorsal side of the segment appears curved forward at the left end. The ducts and pore show clearly the influence of this form. The course of the ducts toward the edge is oblique, *i.e.*, nearly parallel to the dorsal furrows in this region, and the pore lies nearly

in the middle of the edge as it is bounded dorsally. In *b* very similar conditions exist, but the bend in the dorsal surface of the segment is more pronounced than in *a*. The ducts are more oblique than in *a* and elongated, but preserve the same relations to the segment. The dorsal furrow forming the anterior boundary of *b* curves to such a degree that it does not reach the edge at all, thus leaving it apparently undivided, *i.e.*, not distinct from the edge of *c*. Near the middle of this common edge a single pore appears, and into this open the

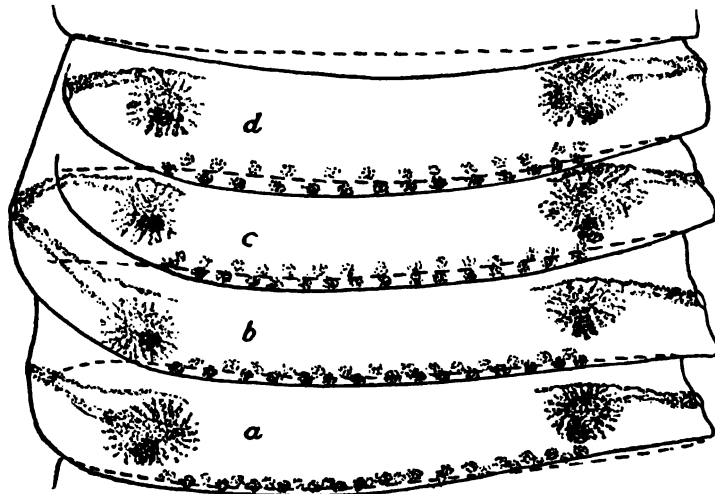


FIG. 37.

ducts from *c* as well as those from *b*. The ducts of the organs in *c* cross the course of the curved dorsal furrow to reach the edge, but this part of the furrow is very slight.

The organs at the left of *d* present an extremely interesting relation with respect to the furrow. The dorsal furrow between *c* and *d* turns anteriorly and runs parallel to the edge, but the furrow in front of *d* is straight. The ducts of the genital organs show no tendency to run parallel to the curved furrow, but meet it almost at right angles, and the pore appears in this furrow instead of upon the edge of the body. The furrow is deeper than the one posterior to it which crosses the ducts in *c*, apparently without affecting their position.

The difference in the relations in these two cases is undoubtedly due to the difference in depth of the two furrows. In the case of *d* the furrow is deep enough either to interrupt the course of the ducts or else to produce conditions approaching those at the edge of the body, and consequently the pore forms here instead of at the edge. The ducts are slightly shorter than the normal, but the inner portions of the organs show a perfectly normal arrangement.

2. *Other Abnormalities.*

Under this head are included a few cases of abnormalities of a different nature from those previously described. Two of these (Figs. 38 and 39) are cases of lateral duplication of the genital organs, and the other two (Figs. 40 and 41) are cases of alteration in position of the genital organs.

Figure 38.

The figure, a view from the ventral side, shows a number of abnormalities. Between the segments *a* and *b* the furrows are normal except near the right edge, where both curve posteriorly. On the ventral surface the furrow ends before reaching the edge, while dorsally it continues to the edge. The inner portions of the genital organs are normal, and the ducts extend in the normal direction toward the edge, but do not reach it. The pore lies on the curved dorsal furrow a short distance from the edge of the body, *i.e.*, on the dorsal surface. The terminal portions of the ducts are entirely normal in structure. Thus the abnormal edge formed by the curved furrow affords conditions which allow a normal pore and terminal organs to appear and so resembles closely the segment *d* in Fig. 37, except that there the furrow between *c* and *d* turns anteriorly instead of posteriorly. At the left of *a* the form of the segment and the organs are normal.

The segments *b* and *c* are incompletely separated on the ventral surface, and *c* is a spiral in consequence of the peculiar curve of the ventral furrow separating the parts *c* and *d*. On

the dorsal surface the furrows are normal, and the regions corresponding to *c* and *d* are parts of a single segment. Thus the spiral furrow bounding *c* and *d* begins ventrally between *b* and *c*, makes one turn, then bends anteriorly, separating *c* and *d*, next makes another almost complete turn and finally ends on the ventral surface anterior to *d*, so that *d* is not completely separated from the segment next anterior to it.

At the right of *b* the edge is very long in consequence of the curve in the posterior furrows, but there are indications that these curved portions of the furrows do not mark the

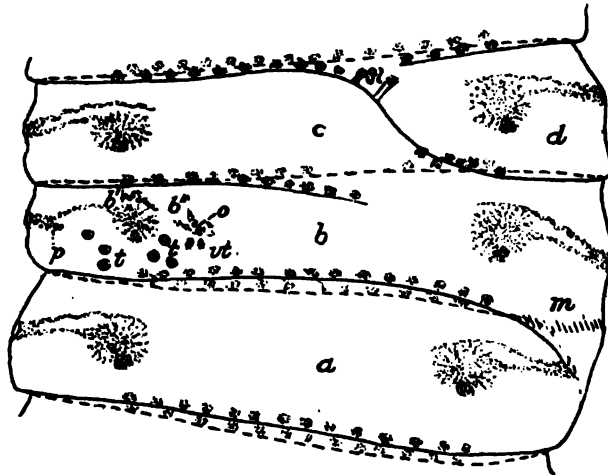


FIG. 38.

actual posterior boundary of *b*. Along the line *m* the cellular structure appears denser and stains more deeply, thus resembling in appearance the regions near the intersegmental furrows. There is no real furrow here, but the presence of this band of tissue similar to that which occurs along segmental boundaries indicates that the posterior boundary of *b* is here and not along the curved furrows; that is, these latter are mere wrinkles in the surface continuous with the intersegmental furrows. If this be the correct interpretation of the conditions here, it is evident that the ovary and pore at the right of *b* present the usual correlation in position with the form of the segment.

At the left of *b* peculiar conditions appear. Besides the organs (*b'*) in the normal position there is another partial set (*b''*) lying to the right of the first. The first set, although normally placed, is imperfect, for the ovary and vitellarium are rather smaller and less branched than usual; the oviduct, atrium, and pore are normal, but the vas deferens is not complete. Its inner end appears anterior to the ovary — near the letter *b'* — but it ends blindly posterior to the oviduct instead of in its normal position anterior to it. A peculiar condition appears in five or six of the testes (*t t*) near the edge. They are enlarged and packed full of spermatozoa, so that they stain like the vas deferens of this stage and are quite different in appearance from the other testes, though the testis cells can be distinguished in them with high powers. The inner portion of the vas deferens is also full of spermatozoa, but the seminal receptacle is empty, indicating that there is no outlet for the sperm into the female organs. In the normal organs of adjacent segments impregnation has already occurred. The accumulation of sperm in the testes (*t t*) is doubtless due to the imperfect development of the male ducts. The movement of the spermatozoa from the middle regions of the segment toward the edges having occurred as far as possible — the testes in the middle region are empty of sperm — they have accumulated in a number of testes near the edge and remain there, since there is no outlet to the exterior or to the female organs. This condition is found in one other case (Fig. 39).

The small size and imperfect development of the set of organs *b'* is probably due to the fact that the left half of the segment *b* is considerably shorter than the normal. The normal length at this age is about that of *a*, and this portion of *b* is only a little more than half as wide as *a*. At the right *b* is wider and normal organs occur.

The second set of organs (*b''*) is very small and rudimentary, consisting of a small simple ovary (*o*) and two small groups of cells representing the vitellarium (*vt*) without any traces of ducts. The orientation of these organs in the proglottid is apparently normal.

This transverse duplication of the female organs does not

appear to be connected with any visible abnormalities in the form of the segment or in the relations of its boundaries. The two sets of organs taken together probably do not represent more material than a single set of normal size. The left side of the segment *b* is somewhat shorter than normal, but in Fig. 39, *c*, where a similar duplication occurs, the segment is of very nearly normal length. The furrows bounding this region of the segment seem to be normal, except that the distribution of the inter-proglottidal glands in the dorsal furrow between *b* and *c* is rather irregular. None appear in this furrow in the middle region of the body, and only a few to the left of the middle. The furrow is normal in appearance, however, and the other furrows seem to be normal in every respect. The conditions found here may perhaps be due to the splitting of a single genital mass in earlier stages, but if this is the case no clue is afforded as to the cause of the splitting. If such a division should occur, later growth would undoubtedly increase the distance between the two portions. From a study of the early stages of the genital organs and their method of origin I am inclined, however, to believe that this extra set has arisen *in situ* and without connection with the set *b'*. If this is the case its appearance must be the result of certain internal conditions, which present no other visible manifestation.

This transverse duplication of organs constitutes a problem entirely different from that of their multiplication longitudinally. Whether or not it is to be regarded as the result of a kind of longitudinal division of the segment is doubtful. No organs except the ovary and vitellarium are duplicated in this case, *i.e.*, the organs on the ventral side only. This is likewise the case in Fig. 39. These two examples are the only ones of this nature which I have found so far, but it is hoped that additional material bearing upon this point may be discovered and may serve to throw some light on the factors concerned in the production of this peculiar abnormality.

The regions *c* and *d* of the ventral side are separated by a portion of the spiral furrow which runs almost longitudinally. At the right it forms the posterior boundary of *d*, but turns anteriorly and then continues as the anterior boundary of *c* at

the left. On the dorsal surface the furrows appear normal. Relations at the two edges are apparently normal, and the genital organs appear normal in all respects.

The relations of the inter-proglottidal glands to the abnormal furrows are interesting. Those portions of the furrow which run transversely show the glands in their usual position, but there are none in the region where the furrow departs from its transverse course. Two of the glands (*gl*) in the partial segment *d* present very peculiar relations to the furrow. Here the furrow bends posteriorly, but the last two glands appear at some little distance anterior to it and almost in line with the others and are connected with the furrow by distinct ducts of considerable length. In the posterior region of the curve two of the glands lie posterior to the furrow as it curves forward, but these open directly. The relation of the glands to this curved furrow affords further evidence in favor of the conclusion that the curved furrows do not always coincide with segmental boundaries. Here the glands do not follow the furrow in this curve, but lie at some distance from it and are entirely absent from that portion which departs farthest from the normal condition. It appears as if the glands follow the line of the real boundary, while the furrow does not. Nevertheless, as the presence of the ducts indicates, the glands tend to open in the furrow.

Figure 39.

The series of abnormalities occurring in the three segments represented here is in some respects the most peculiar that I have found in this species. The figure is a ventral view.

In the segment *a* the relations on the right are normal, but on the left there appears on the dorsal surface the anterior end of a spiral, the remainder of which is not drawn, as it makes only one turn and is similar to others already discussed. The ducts of the organs *a'* extend to the surface in accordance with the relations on the dorsal surface and thus open at a point on the edge which is dorsally a part of *a*, but ventrally in another proglottid.

The segment *b* is abnormally short, even more so dorsally than on the ventral surface. In accordance with this fact only partial organs are developed right and left (*lb'* and *rb'*). Ducts connecting with the surface do not appear at all, and on the left no pore is formed. On the right edge, however, a pore appears, of normal size and with atrium and a small portion of the oviduct extending inward from it. This portion was found upon examination to present the characteristic appearance of the oviduct and to possess a lumen, but was

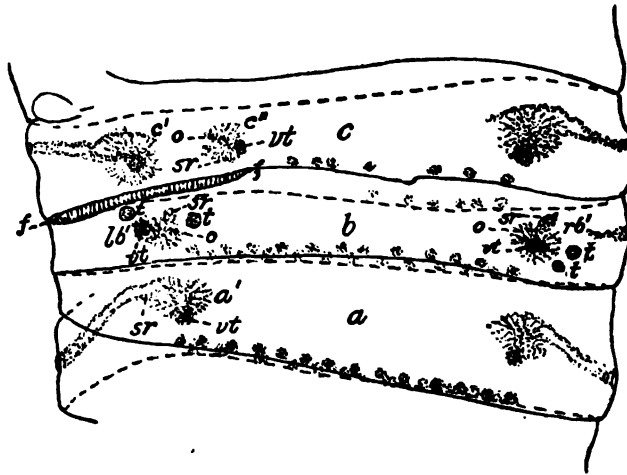


FIG. 39.

closed at the inner end. The reason for the appearance of a pore on the right edge and not on the left lies, I believe, in the fact that the right edge is longer than the left and thus presents more nearly the conditions of the normal edge. Each of the partial female organs *lb'* and *rb'* consists of a small ovary (*o*), a small vitellarium (*vt*), and the inner portion of the duct, terminating in a small, bladder-like, closed seminal receptacle (*sr*), which is empty. The organs of the dorsal surface are less nearly normal than those of the ventral. Testes appear, but their number is much less than the normal, even in proportion to the small size of the proglottid, for they are scattered very sparsely through the middle region, while other segments of this stage show large numbers of them. No traces of vasa

deferentia appear on either side. This rudimentary condition of the male organs is doubtless due to the extreme shortness of the proglottid. As on the left side of *b* in Fig. 38 the spermatozoa have accumulated in some of the testes in the lateral regions of the segment (*t t*). In this case there is no exit on either side of the spermatozoa, and the female ducts are incomplete and unconnected with the male ducts. Consequently the segment is not functional. The spermatozoa cannot fertilize the eggs of this or any other segment, and the eggs cannot be fertilized by spermatozoa from this segment or from any other.

The ventral furrow between *b* and *c* is distinctly abnormal, especially at the left. A normal furrow does not cut into the body vertically, but obliquely, and in such a manner that the posterior edge of each proglottid seems to overlap the anterior edge of the next succeeding. Over about two-thirds of its course the ventral furrow between *b* and *c* is normal in its relations, though slightly irregular in its course. Over the remaining third, however, — the shaded portion at the left marked *f*, — it is a vertical furrow widely open to the surface, and nearly twice as deep as the normal furrow by actual measurement. It cuts almost halfway through the body and thus separates *b* and *c* in this region much more completely than they are separated elsewhere. This portion of the furrow shows no inter-proglottidal glands, but they are present in the more nearly normal portion. The ventral furrow anterior to *c* is also abnormal at its left end. It is interrupted, one portion turning anteriorly, and the other curving near the edge so as nearly to enclose a small area. The dorsal furrow bounding *c* is normal.

In the segment *c* there are two normal sets of organs, the one situated normally, and the other nearly so ; but in addition to these organs a third set (*c''*) appears situated to the right of *c'* and consisting of a small ovary (*o*), a vitellarium (*vt*), and a small, empty seminal receptacle, which is closed. This case of transverse duplication of organs is very similar to the one figured in *b* in Fig. 38, and it is in the same region of the body, the two being separated by some thirty segments only. This second case does not afford any evidence as to the factors concerned

in the production of this form of variation. However, the position of the organs in *b* and *c* does bring to light some interesting facts regarding the orientation of the organs in the segment, indicating that this also is perhaps correlated with the "form" of the segment.

In the normal form of the female organs the vitellarium lies more or less completely posterior to the ovary, as is clear from many of the figures (see *a'* in segment *a* of this figure, for instance). The seminal receptacle appears at a point separated from the vitellarium by one-quarter of the circumference of the ovary, *i.e.*, about ninety degrees (note the position of *vt* and *sr* in *a'*, where they are normally situated). Now in the supplementary set of organs *c''*, in the segment *c* the vitellarium lies on the right side of the ovary, while the small rudimentary seminal receptacle (*sr*) is posterior. That is, the whole set of organs appears as if rotated through an angle of ninety degrees from its normal position. An oviduct, if present and normally oriented with respect to the ovary, etc., would lead to the shaded portion of the furrow *f*.

Turning now to the organs *lb'* at the left side of *b*, we find that the parts present in this set are the same as those found in *c''*, *viz.*, a small ovary (*o*), a vitellarium (*vt*), and a small closed seminal receptacle (*sr*). The orientation of this group, however, is different from that of *c''*. The vitellarium, instead of being in its normal position posterior to the ovary, lies at the left of it, while the small seminal receptacle (*sr*) is anterior to the ovary, instead of to the left. Here, then, the whole complex appears as if rotated through an angle of ninety degrees in the direction opposite to that in which the rotation of *c''* is conceived as having occurred. In consequence of this position, an oviduct, if present and oriented normally with respect to the other organs, would, as in the case of *c''*, open into the shaded portion of the furrow *f*.

Examination of the organs *rb'* at the right of the segment *b* shows that the relations there are more nearly normal. The vitellarium (*vt*) is somewhat posterior to the ovary, and the seminal receptacle, *sr*, though somewhat more than ninety degrees from the vitellarium, does extend in a nearly normal

direction, and if growth of the parts continued, the receptacle and the oviduct from the pore would meet. In brief, this set of organs shows what is practically the normal orientation, while lb' and c'' do not.

The suggestion which offers itself in this connection is that the abnormal orientation of the organs c'' and lb' is correlated with the presence of the furrow which lies between them. The furrow is on the ventral side, as are these organs. May it not be possible that the orientation of these two sets of organs with respect to this furrow is due to its extreme depth? They are oriented with regard to it as if it were the edge of the body, though no pores appear, opening into it. There is a considerable extent of free surface in the dorso-ventral plane on the sides of the furrow. The organs at c'' are far from the real edge, but near this abnormal furrow, and their abnormal orientation appears to be a form of adaptation to the abnormal conditions. The complex of organs lb' , as described, is correspondingly oriented with regard to the furrow f . The question at once arises as to why this should be, since this set lies at the normal distance from the real edge. The answer to this question may lie in the fact that the left edge of the segment b and the region near it are very short — apparently too short to allow a pore and ducts to appear. The organs at lb' are much nearer to the furrow f than to the edge, and if the orientation of c'' is determined by the furrow, that of lb' may be also. In the organs c' orientation and position are apparently normal. The pore lies rather far anteriorly, and this is probably due to the fact that c is incompletely separated dorsally from the segment next in front. The position and orientation of this set of organs may seem to be strong evidence against the conclusion that the orientation of c'' and lb' is due to the presence of the furrow f ; but here the edge is normal and the proglottid is of normal length, *i.e.*, normal relations are possible here, while they are not in the cases of lb' and c'' . Furthermore, on the right of b , where the furrows are both normal, and the edge is wider than at the left, the organs are normally oriented and a pore is present, though the parts are not connected.

All who study the cases discussed in this and the preceding paper must, I believe, conclude that the position and arrangement of the genital organs in abnormal as well as in normal proglottids are very definitely determined by the form relations

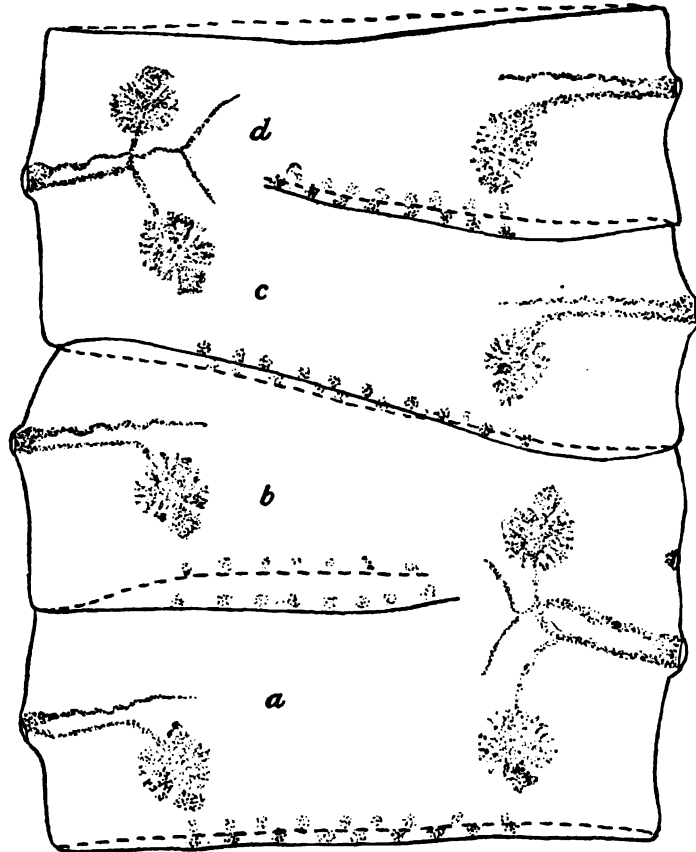


FIG. 40.

of their segment. The above suggestions have been made in the belief that the form relations may have some influence here, and in the hope of throwing some little light on the conditions in this particular case. Whether other similar cases, if found, will confirm them remains to be seen.

Figure 40.

Two cases of partial division are figured here as seen from the dorsal surface. The two are almost exactly similar, except that in one case the partial furrows extend from the left edge, in the other from the right. At the side of the body, which is completely divided in each case, two complete sets of genital organs normally situated are found. At the other side the inner portions are double, but the ducts unite to form a single

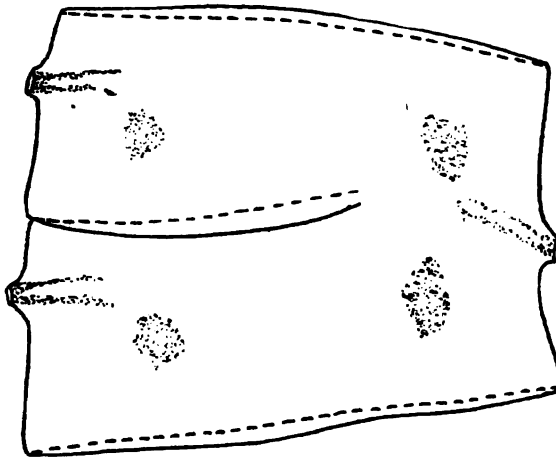


FIG. 41.

vas deferens and a single oviduct, and these open through a single pore, which is situated almost exactly in the middle of the undivided edge. The most remarkable fact is that the position of the anterior set is the reverse of that of the posterior set in each case, the oviduct running posteriorly from the ovary, and the vitellarium lying anterior instead of posterior to the ovary. The vas deferens also runs posteriorly instead of laterally. The arrangement of the organs is very evidently correlated with the incomplete separation of *a* and *b* and of *c* and *d*. The undivided edges of *a b* and *c d* are shorter than the divided edges, but the undivided edge of *a b* is longer than the undivided edge of *c d*, and it is interesting to note that in the region corresponding to *b* of the edge *a b* a second pore appears,

much smaller than the one posterior to it and without any ducts opening into it. Apparently the greater length of this edge has afforded space, as it were, for the formation of another pore, but the ducts connecting with it fail to appear. This case, like a number of others, shows that the pore may be wholly unconnected with the other organs, but that its formation is doubtless due to the general conditions that lead to the formation of the other portions of the set, even though the two parts do not unite.

Figure 41.

This case is very similar to the one shown in Fig. 40. The development is so far advanced that it is impossible to determine with certainty whether the division of the ducts is as complete as in that case. The cell-masses at the right representing the two ovaries and vitellaria appear about equal in size, and there are indications that they were connected with the single duct leading to the pore. It is impossible to determine whether the vas deferens divides or not.

HULL ZOÖLOGICAL LABORATORY,
UNIVERSITY OF CHICAGO,
May, 1900.

A CONTRIBUTION TO THE DEVELOPMENT OF PARYPHA CROCEA.¹

CARRIE M. ALLEN.

My work has been carried on at the Zoölogical Laboratory of Syracuse University under the direction of Dr. C. W. Hargitt, to whom it is a pleasure to express my obligations for his kind suggestions and supervision throughout its progress, and for the pains which he has taken in furnishing me with the best of appliances and material. The material upon which the investigations have been made was collected by Dr. Hargitt at Woods Holl, Mass., during the summer of 1898. Through his kindness I had at my disposal an almost limitless supply killed and preserved by a number of methods. Picro-sulphuric acid and Perenyi's fluid both gave excellent results, but formalin proved unsatisfactory for histological work.

In most of my study I used preparations stained *in toto* in borax-carmine. The specimens were left in alcoholic borax-carmine for twelve hours, after which the stain was extracted by acid alcohol for from fifteen minutes to half an hour. In dehydrating they were left in each of the various grades of alcohol for thirty minutes, after which they were cleared either in cedar oil or chloroform. Both clearing agents gave good results, but the latter was preferable because of its rapid action.

In staining on the slide, iron-haematoxylin and double stain of eosin and haematoxylin both proved satisfactory.

In using the iron-haematoxylin the sections were fixed to the slide, carried down through the alcohols to 50 per cent, and placed in a 2 per cent solution of ammonio-ferric-alum for from thirty minutes to three hours. They were then washed in running water for twenty minutes, stained in 0.5 per cent aqueous solution of haematoxylin for from one-half hour to

¹ *Contributions from the Zoölogical Laboratory, Syracuse University.*

two hours, washed again in running water and cleared for a few seconds in iron-alum. After rinsing in distilled water they were plunged into 95 per cent alcohol, carried up and mounted in balsam. The best results were obtained by leaving the slides in each stain for an hour. This method was of greatest value in study of the segmentation of the egg. The cytoplasm appeared gray, chromatin fibers black.

In using the eosin-haematoxylin method the sections were stained for an hour in a 2 per cent solution of eosin in 90 per cent of alcohol, after which they were stained in a weak solution of Delafield's haematoxylin for twenty minutes. This stain gave very good general differentiation, and was of especial value in determining the origin of the sex cells.

A number of other stains and combinations of stains were used with fair success.

General Description of Parypha.

Parypha crocea is found all along the New England coast attached to floating timbers and the piles of wharves. It seems to prefer brackish water and partial sunlight, but often occurs in pure sea water.

This hydroid grows in colonies which arise from a single individual by a process of budding, and the sexes are always separate. The hydrorhiza is made up of a contorted mass of irregularly branched stems, from which the hydrocaulus of the individual hydroid arises. The stems bearing the adult polyps are usually two and a half to three inches in length, and short stems are sometimes found branching out from the main ones. Enclosing the stem is a horny, often annulated sheath, the perisarc. The polyp is borne at the top of a somewhat globular expansion of the hydrocaulus, and is almost conical in form, with a broad, saucer-shaped base. It contracts about three-quarters of the way up, forming a thick-walled, flexible proboscis, in the center of which lies the mouth, surrounded by a circle of short, thick tentacles with decurrent bases. Around the base of the polyp is a circle of long, slender tentacles, varying in number from sixteen to twenty-four. The medusoids

are borne upon long, slender, branched peduncles which arise a short distance above the tentacles of the lower row. All parts of the hydroid are made up of the two layers characteristic of all hydroids, but the mesogloea forms only a thin layer in the peduncles and tentacles and is not visible in the medusoids. The tentacles of both rows consist of a central axis of the endoderm, surrounded by a thin layer of ectoderm.

Origin and Morphology of Male Gonophores.

The medusoids in this species begin to appear early in the development of the hydroid, when the head upon which they are borne is less than a quarter of the size of the adult polyp. The first indication of their formation is a slight outpushing of the endoderm of the body wall a little above the axils of the lower row of tentacles. The ectoderm is pushed out and becomes thinner than in the adjacent parts of the wall. The papilla thus formed elongates into a peduncle communicating directly with the body-cavity of the polyp. From this peduncle arise short branches which may subdivide, and it is at the ends of these that the medusoids are borne. At first there is merely a thickened layer of endoderm surrounded by a thin layer of ectoderm, but when the length of the bud is about once and a half the width, the ectoderm cells at the tip begin to grow rapidly, forming a plug of cells with large nuclei and indistinct boundaries (Pl. I, Fig. 1). For a time the endoderm is forced back (Pl. I, Fig. 2), but it soon begins to grow down into the center of the plug, to form the manubrium, and around the outside to form the endodermal layer of the bell. All the cells between this layer and the manubrium are of ectodermal origin, and from them the reproductive elements arise. The sex cells increase in number, and to some extent in size, until they occupy the greater part of the bell. While this growth is taking place the cells at the distal end of the gonophore next the endodermal layer of the bell begin to differentiate, forming a thin, delicate layer which gradually extends around the gonophore and becomes the inner wall of the bell (Pl. I, Fig. 3). It is made up of cells much smaller than those from which they

are derived. Meanwhile the endodermal layer has thickened in four regions equidistant from each other at the distal end of the medusoid. At first the cells in the thickened region are irregularly arranged, but later they form themselves into two rows with a space between them. In a few cases no cavity was found, and in two of the gonophores examined it extended halfway around the bell. The remainder of the wall consisted of a single layer. Pl. I, Fig. 4, represents the condition found in most of the medusoids. Agassiz ('62), p. 259, states that there are neither radial nor circumoral canals in this species but the position and mode of development of these cavities leave little doubt that they are rudimentary radial canals. No circular canals were observed, and the radial canals were never found connected with the body cavity of the medusoid in any of the hundreds of sections studied.

Spermatogenesis.

The large nucleated cells lying between the manubrium and the inner wall of the bell become the sperm mother-cells, which finally break up to form the sperms. In the first division the karyokinetic figures are distinct and show spindles and prominent chromosomes. The later stages were difficult to study because of the minuteness of parts, and I was unable to demonstrate clearly the exact number of spermatozoa derived from a single germ cell, but I think four are usually formed. Their structure could only be made out in particularly favorable sections, but was easily demonstrated by crushing the gonophore and allowing the sperms to escape. They consist of a pear-shaped head with a very long, slender tailpiece. When fully developed the male gonophores are spherical, and the walls are so thin that their structure can only be determined by the use of very high powers. They bear no tentacles, although the ectoderm is sometimes thickened slightly in the regions where tentacles arise in the female. I examined carefully a large number of mature male gonophores to learn whether or not the ectodermal layer of the manubrium was formed and discovered a definite transparent layer next the

endoderm. No gonophores from which the sperms had been expelled were found, so I was unable to prove that this represented the ectoderm of the manubrium, but as that layer was not observed in the female until a very late stage, I think that there can be little doubt that it functions as such.

Origin and Morphology of the Female Gonophore.

The female gonophores arise in the same manner as the male and, in the early stages, are made up of the same parts, but later may always be distinguished by a circle of six or eight short, blunt tentacles at the distal end and by their more elongated shape. When filled with young they are nearly spherical and the tentacles mere papillae; but when the larvae have been set free the medusoids become elongate and the tentacles expand. Pl. I, Fig. 5, shows a section through two of these tentacles.

Oögenesis.

The primitive egg cells are developed in the same manner as the sperm mother-cells. I find no evidence whatever of ova either in the coenosarc of the stem, the body of the polyp, or the walls of the peduncle. There are in the endoderm of the polyp and peduncle numerous large, deeply stained cells with large nuclei which somewhat resemble eggs when cut in the right plane, but a careful study of a large number of sections reveals the fact that they are in reality highly differentiated endodermal cells. They are always in contact with the supporting layer and usually project beyond the other endoderm cells into the body cavity, neither of which conditions, according to Weismann ('83), p. 70, occurs in egg cells. Moreover, these cells are much larger than the primitive ova and take a deeper stain than do the eggs in any stage of their development. They are very rich in protoplasm, and sometimes the outer surface is found sloughing off into the body cavity. This condition was even more marked in similar cells in *Eudendrium ramosum*, where they extend farther into the body cavity, and the discharge of portions of their protoplasm was

very evident. All this would indicate that they were glandular in function. They are largest and most numerous in the peduncle and occasionally one is found in the manubrium, but such cases are rare. It would be impossible to distinguish a section through the peduncle of a male head from that of a female, as these cells are equally conspicuous in both. Pl. I, Fig. 8, shows a number of these glands, one of which resembles an egg, but other sections through the same peduncle show that it is really in contact with the supporting lamella.

In the younger stages of development the manubrium of the female appears to consist entirely of endoderm, but when the gonophore is fully mature and the primitive ova have disappeared, a thin layer of ectoderm is found to be present. It consists of a single layer of much flattened cells with smaller nuclei than those of the germ-tissue cells from which they are derived.

Development of the Ovum.

The primitive egg cells make up the large mass of tissue lying between the manubrium and the inner wall of the bell. They are packed closely together, so that the outlines of the cells are more or less irregular. The nuclei are large and spherical and contain a prominent nucleolus which takes a very deep stain. The mass of protoplasm surrounding each nucleus is small, and the cell boundaries are very indistinct (Pl. I, Fig. 4).

As the gonophore grows older the nuclei of the germinal tissue become much larger and more prominent, the mass of protoplasm surrounding them increases in bulk, and the cell boundaries become more clearly defined. At this stage the nuclei appear as very large spheres, with the chromatin fibers arranged in a sort of network around the periphery. Within the layer of chromatin is a colorless mass, near the center of which lies the nucleus suspended by four or five slender threads, which run out to the layer of chromatin. These threads take a fainter stain than the chromatin fibers and are only visible in especially well prepared specimens.

The nucleolus is usually spherical or slightly elongated,

but in many cases it shows a varying number of short, blunt processes. This condition was most clearly seen in sections stained with ammonio-ferric-alum and haematoxylin.

Within the nucleolus are a number of small, transparent, highly refractile bodies, the nature of which will be discussed later. There is usually one of these in the nucleus of each primitive ovum, but some contain two. As the ova grow older the number increases and there are sometimes as many as four or five in a single nucleolus. The protoplasm of the cells is granular and often contains a few small vacuoles.

Up to this time the growth of the various cells of the germinal tissue has been about equal, but now several cells increase markedly in size, and often the greater number in one side of the gonophore are found to be thus growing. If, however, a large number take part in this early development, the cells in the opposite side of the gonophore decrease in size, both nucleus and cytoplasm becoming smaller. Soon a few cells attain greater size than the rest and develop very rapidly. Many of the cells in this and the preceding stages are found to possess pseudopodia-like processes quite similar to those figured by Doflein ('96) for *Tubularia*. Smallwood ('99) mentions the same condition in the eggs of *Pennaria*. The pseudopodia extend in between the other primitive egg cells, and the tips are more granular and take a deeper stain than the rest of the egg. Doflein ('96) has given much attention to the amoeboid forms assumed by eggs of *Tubularia*, and he inclines to the belief that these processes do not function as mouths by which the surrounding eggs are bodily engulfed. My results have coincided very closely, in most respects, with those of Doflein, but numerous cases were also observed where the outline of the absorbed egg could be definitely made out within the protoplasm of the absorbing egg. Even in these cases, however, the absorbed egg did not lie in a vacuole, as would the food taken in by the amoeba, and the outline could only be made out by the greater density of its protoplasm. It seems, therefore, that in this case also we have a blending of the protoplasm of the two cells rather than a digestion and absorption of the one by the other. There seems to be no great uniformity either

in the number or location of the primitive cells which finally become ova, although by far the greater number lie next the manubrium, and few, if any, develop on the outer surface of the germinal mass. I am inclined to agree with Doflein ('96), p. 65, that all of the cells of the germinal tissue have potentially the capacity of becoming eggs, but that those favored by better nourishment or advantage of position are the first to develop.

He says in this connection : "Das starke Wachstum des Gonophors hat einzelne Lücken und Spalten im Gewebe entstehen lassen, und in diese wachsen nun die Keimgewebezellen mit ihren Fortsätzen hinein." But while in my investigations many such cracks were found, in most instances the pseudopodia extended between eggs where no crack occurred, and in the greater number of ova no pseudopodia were present at all. I am, therefore, led to the belief that proximity to cracks in the germinal tissue is not of controlling importance, although the eggs do undoubtedly take advantage of the room afforded by such cracks when present. Doflein ('96) also states that in *Tubularia* the growing eggs are always found next the manubrium or upon the outside of the germinal mass, unless cracks are present within the tissue. I have examined a large number of sections, and I find that in *Parypha* the eggs of the outer layer are the last to develop, but that those in the interior of the germinal tissue are often found considerably enlarged even in the younger gonophores.

When the growing cells have attained a diameter about three or four times that of the cells of the germinal tissue, the nucleus is found lying close to the periphery of the egg and is oval and transparent, the chromatin fibers being scarcely visible (Pl. II, Figs. 5, 6). The nucleolus takes a fainter stain, and in most cases contains a number of the refractile bodies already mentioned. Later these bodies apparently unite, as nearly the whole nucleus is often occupied by a single large one. Just what their character is I am unable to state, but they appear to contain oil, and certainly they are associated with the peculiar metabolism exhibited by the cell at this time (Pl. II, Figs. 5-7). In some of the eggs in which the nucleus had this peripheral position, its outline was irregular upon the

inner side so that it resembled the figure shown by Hickson ('90) to illustrate the stage in the fragmentation of the oöperm nucleus of *Allopora*. In the next stage the nuclear membrane is broken down and the nucleoplasm blends with the cytoplasm of the egg, from which it can only be distinguished by its homogeneousness and greater transparency (Pl. II, Figs. 7, 8). In other eggs having the same general appearance as the last no nucleus whatever is visible. I have several complete series through eggs in this stage, none of which show any signs of a nucleus, although they have been stained by a number of different processes, and I am perfectly confident that the nucleus would be visible if present. Hickson describes a similar condition in the eggs of *Allopora*, *Millepora*, and *Distichopora*; and Dr. C. W. Hargitt tells me that in his opinion a like condition is to be found in *Eudendrium*, although he has not yet placed it beyond doubt.

Hickson ('93) has written an extended account of "nuclear fragmentation," in which he cites the opinion of a number of authors with regard to this much disputed question. After describing the stages observed in *Distichopora* he says: "I have described a process which can only be compared with the so-called free nuclear formation in the early insect embryos. Nuclei make their appearance in places which were previously devoid of any nucleus or nuclear structure. It is not reasonable, however, to assume on the insufficient evidence before us that "nuclear formation" does actually occur. It seems to me much more probable that minute fragments of nuclear substance scattered through the protoplasmic meshwork collect together in places, and form by their fusion true recognizable nuclei. In other words, the process we have under observation is rather one of "nuclear regeneration" than one of free "nuclear formation." He quotes Flemming and Ziegler as authorities most opposed to this view, both these investigators contending that any process of nuclear division other than that by mitosis is a sign of the degeneration of the nucleus and the approaching end of the life of the cell. Ziegler inclines to the opinion that nuclei which have arisen by amitotic division will never again divide mitotically. Opposed to these are the works

of Verson, Frenzel, Löwit, and others who, since the publication of Ziegler's paper, have called attention to cases of amitotic division of the nucleus which are certainly not followed either by nuclear degeneration or a cessation of cell multiplication. Altogether there seems to be constantly increasing evidence that such a fragmentation does occur in the ova of widely separate groups of animals.

Wilson ('96), p. 85, believes that the subject requires more study, but says: "There can be no doubt, however, that Flemming's hypothesis in a general way represents the truth, and that in the majority of cases amitosis is a secondary process which does not fall in the generative series of cell division."

Absorption.

At about the time when the transparent nucleus lies near the periphery of the egg the cytoplasm changes from a granular to a reticular structure. The boundaries between the large cells and those adjacent to them now begin to break down and the protoplasm to blend. This fusion may take place between two large cells or between a growing cell and a germ tissue cell. The former usually occurs first, the large cells near the manubrium fusing and then gradually taking in the germ cells which surround them. The nuclei of the latter are found lying in the protoplasm of the absorbing cell. Both conditions are shown in Pl. I, Figs. 6, 7.

The outline of the syncytium thus formed is very irregular, and parts of the walls of the constituent cells persist for a time, showing where the fusion has occurred (Fig. 7). Doflein ('96), p. 66, states that in *Tubularia* one large, well-nourished cell controls the absorption, and that as it grows its nucleus also increases in volume, and that the nucleus becomes the functional nucleus of the ovum, the other nuclei being gradually absorbed. In *Parypha*, as already stated, the nuclei of the growing cells disappear at an early stage so that only the nuclei of the smaller cells persist. It thus becomes impossible to tell which is the controlling cell.

Finally the mass of fused cells takes on the typical egg

form, the protoplasm near the periphery becomes more dense, and the absorbed nuclei are found in various stages of disintegration. The egg now lies on the outside of the mass of germinal tissue and next to the wall of the bell. No evidence of fusion with the primitive eggs was observed after this stage was reached, although the two were still in contact. It is quite evident, however, that the remaining germ cells grow and unite to form new eggs later in the history of the parent, since primitive eggs are often found in advanced stages of growth, while two or three nearly mature embryos still occupy the gonophore. In other cases the gonophores contained several embryos in various stages of development, but no primitive ova. Doflein ('96), p. 67, states that, although he was unable to obtain sections to illustrate adequately the point, he believes that the germ cells of *Tubularia* do unite to form new eggs after the larvae have left the gonophore. In *Parypha* there is no chance to doubt that new ova are formed even before the exit of the larvae.

Fertilization.

As is the case in many of the hydroids, the process of fertilization is shrouded in mystery. The fact that the eggs are developed in closed gonophores makes it difficult to decide just when fertilization takes place. In discussing the development of *Allopora*, Hickson states that he believes that fertilization occurs while the nucleus lies at the periphery of the egg, and previous to the time when it becomes irregular in outline. From the positions of the eggs in which these irregular nuclei were found, *i.e.*, next the manubrium, this might be the case here, but nothing was discovered which threw any light directly upon the matter.

History of the Pseudo-Cells.

The nuclei of the absorbed cells are found in various stages of disintegration within the ovum. Some of them resemble so closely the nuclei of the germ-tissue cells that, were it not for the position and the vacuoles within which they lie, it would be

impossible to distinguish them. Later the chromatin fibers lose their reticular arrangement and assemble into a varying number of small spheres just within the periphery of the nucleus, and at the same time the threads which support the nucleolus disappear. The ground material in which the chromatin is suspended, and which up to this time has been nearly transparent, now begins to react to the staining agents, and the structure of the nuclei becomes obscure. If, however, methyl-blue was used, this substance was only slightly affected, so that this stain proved most satisfactory for the study of the various phases exhibited by the retrograding nuclei, or pseudo-cells, as they are sometimes called. Many of the nuclei are often found in the process of division. The nucleolus lengthens slightly, and finally separates into two parts. Later the entire nucleus divides and part of the chromatin goes with each half. Cases in which the nucleolus had divided were very numerous, but very few were found in which the division was actually taking place. Pl. II, Fig. 11, shows such a one, and Fig. 10 represents a nucleus in which there were three processes on the nucleolus. No chromatin fibers were visible in either of these cases. The halves thus formed often divided again, sometimes before they were separated, and in some instances as many as six parts can be observed. The chromatin globules vary in number and size in the various parts (Fig. 12). In some of the nuclei the division is less regular, and portions are often found in the process of being absorbed into the protoplasm of the egg. Fig. 15 represents a nucleus in which the parts formed by the first division were of very unequal size. In the smaller the nucleolus has again divided, but the larger part has been partially absorbed. Often several of these nuclei are found in a single vacuole. Fig. 9 shows one in which there were seven in various stages of disintegration, but usually not so many are found. Doflein believes that they are carried into the vacuoles by currents in the protoplasm. All this goes to strengthen the opinion of Doflein that the absorbed nuclei take the place of the yolk-granules, which are wanting in this species, and that they are gradually broken down to serve as food for the developing egg. They persist through the entire

embryological development, being very numerous in the endoderm of the young hydroid when it escapes from the gonophore. Isolated ones are even found in the endoderm of the tentacles, as noted by Doflein, but I cannot agree with him that they are entirely confined to that layer.

Segmentation of the Ovum.

The egg, after assuming the typical form already described, goes into a resting stage, as a large number are found in that condition and without nuclei. Soon, however, an irregular mass of nuclear matter appears at one pole. Sometimes this forms a single mass, in other cases it is made up of two or three more or less isolated portions. Whether these are finally assembled to form a single nucleus, or whether two or three nuclei are thus produced, I am unable to say, as many of the sections in the later stages might be interpreted either way. In some of the eggs a single definite star-shaped nucleus was present, but in others there were two, and in one case four of these nuclei lying close together at one pole of the egg. There was nothing in these eggs to indicate that the nuclei had not been derived from a single nucleus, but, on the other hand, some of the disorganized masses of nucleoplasm could not but give the impression that more than one would be formed. However, the number is of minor importance, and the real interest attaches to the fact that such a reorganization occurs at all. That it does, I am fully convinced. I have examined a large number of sections with this question particularly in my mind, and am forced to the conclusion that the nucleus of the mature egg is formed by the reorganization of the fragments of the nuclear matter scattered through the cytoplasm.

The earliest stage in which definite mitosis was observed was in the egg shown in Pl. III, Fig. 1. In this three definite nuclei, one in a process of division, showed in a single section. Another section through the same egg revealed a fourth nucleus which, from its position, might have been derived from one of the others, but no spindles were observed. There were no signs whatever of segmentation planes in this egg. The development in the eggs of *Parypha* is very irregular indeed,

and seems to be governed by no single law. In some cases definite cell walls were found in the earlier stages, as in Fig. 2, where four cells had been formed, one of which contained two nuclei. In this we have only the stage next to the one last described, but in that there were no segmentation planes at all. In still later stages the development is quite as irregular. Fig. 5 shows a section in which six nuclei were visible, and other sections through the same egg contained several others, some of which were in the process of division, but no cell walls had been formed. In Figs. 6 and 7 we have sections through much older eggs, but the same indefiniteness of structure prevails. From the foregoing illustrations it will be seen at once that there is little uniformity in the early development of the eggs of *Parypha*, either as to size of the cells formed or the number of nuclei that appear previous to the formation of the cell walls. Segmentation does, however, begin at one pole, and the greater part of the egg is for a time unsegmented. In no case did I find the egg divided into two equal parts, as Dr. Hargitt has sometimes observed in *Pennaria* eggs. Pl. III, Fig. 3, represents conditions similar to what is constantly met with in eggs of *Pennaria*. In total segmentation the ovum consists of a solid sphere of cells of more or less uniform size but with irregular outlines. They are very reticular in structure, and large vacuoles are numerous.

Formation of the Ectoderm.

Following the complete segmentation, the first indication of a differentiation into ectoderm was observed in an increased amount of cytoplasm in the outer layer of cells. These cells then divide radially, forming narrow cells, as shown in text Fig. 1. The two mitotic figures lay in adjacent cells, as shown in the drawing. In the next stage observed the ectoderm appeared to consist of two layers of cells much smaller than those of the endoderm, and distinguished from them by the greater density of protoplasm. The two layers appeared in this case to be dovetailed into each other, as shown in Fig. 2 of the text. In the fully formed ectoderm the cells are very elongate and somewhat

spindle-shaped, with one end broader than the other. Both the form of the cells and the position of the nuclei indicate that they have been formed from a condition like that in Fig. 2, and not by further delamination of the outside layer alone. At this stage the larva is made up of a solid mass of irregular cells with spherical nuclei surrounded by a single layer of much elongated cells. No segmentation cavity is formed. The origin of the germinal layers agrees, therefore, quite closely with that described by Hickson ('93) under *E. e.*, p. 52: "A sterula is formed by precocious delamination. No segmentation cavity is formed, and segmentation is at first incomplete."

Parypha is not mentioned by Hickson under this class, and Tubularia, the form most like Parypha in its general mode of development, he includes under another head. Dr. Hargitt informs me that nothing equivalent to true delamination or invagination occurs in Pennaria. It would, therefore, seem that no one, two, or even three laws of cleavage are sufficient to explain the varied conditions to be found in the segmentation of the hydroid egg.

The embryo now appears concave upon the side next the manubrium, but this is probably due to pressure and not to any intrinsic cause. After the formation of the ectoderm the two layers of the embryo evaginate at seven points so that a section through the region of the process appears star-shaped,

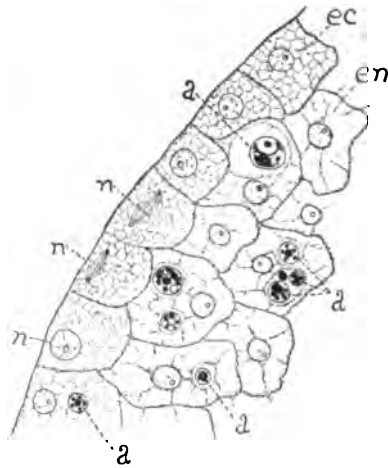


FIG. 1 x 560. — Early stage in the formation of the ectoderm of the embryo; *ec*, ectoderm; *en*, endoderm; *n*, nuclei; *n¹*, nuclei in the process of division.



FIG. 2 x 560. — Later stage in the formation of the ectoderm; *ec*, ectoderm; *en*, endoderm; *a*, nuclei of absorbed cells.

the rays at first being very short. These processes elongate to form the basal tentacles of the young hydroid. While this growth is taking place the convex side of the embryo becomes still more convex, and the concave portion between the tentacles evaginates and becomes convex also. In this way the endoderm cells in the center are split apart and the body cavity is formed. At first it is very irregular, but later the endoderm cells assume the typical endodermal form and arrange themselves in a single layer within the ectoderm, and the body cavity

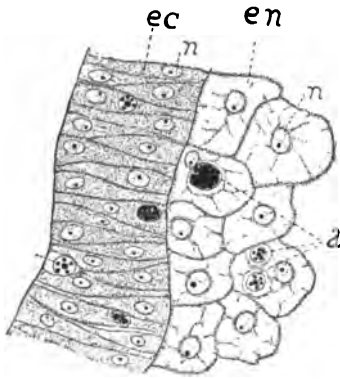


FIG. 3.

FIG. 3 $\times$ 560. — Fully formed ectoderm from convex side of embryo; *ec*, ectoderm; *en*, endoderm; *n*, nuclei; *a*, nuclei of absorbed eggs.

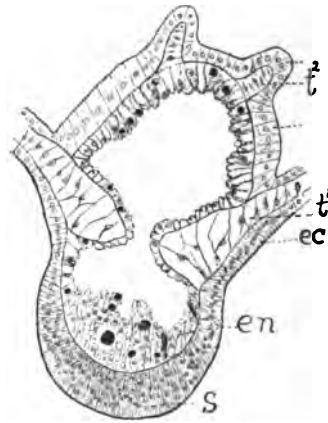


FIG. 4.

FIG. 4 $\times$ 190. — Young embryo ready to escape; *t*, basal tentacle; *t'*, buccal tentacle; *s*, stalk; *ec*, ectoderm; *en*, endoderm.

takes on a form quite similar to that of a young polyp. From almost the earliest stage in the development of the ectoderm the cells on the convex side of the embryo appear much longer than upon the opposite side, and it is this portion which becomes the stem to which the young hydroid attaches itself. According to Agassiz the larva escapes in this condition, and the mouth and buccal tentacles are developed after it attaches itself. I have, however, obtained sections of a large number of mature larvae in which well-developed tentacles were present. Fig. 4 represents an embryo that was just leaving the gonophore. The body and the stem were both well developed, and the basal tentacles were nearly twice as long as the body.

At the buccal end there were five or six tentacles, a section of which is shown in the figure. Whether the mouth is developed at this time or later I did not decide.

Summary and Conclusion.

In a summary of the results obtained in this study, the following points should be noted :

1. The medusoid develops from a bud formed by an outgrowth of the body wall and shows itself first in a thickening of the endoderm.
2. The sex cells in both the male and the female are derived from the plug of ectodermal cells which is formed at the apex of the bud.
3. The medusoid is never set free and no circular canal is formed, although remnants of four radial canals are quite conspicuous.
4. The eggs grow by the absorption of the cells of the germinal tissue, a syncytium being thus formed.
5. The nuclei of the primitive eggs persist as pseudo-cells and are gradually broken down to serve as food for the growing embryos.
6. The pseudo-cells divide amitotically, but are finally absorbed by the growing egg.
7. The nucleus of the growing egg is absorbed at an early stage, but is re-formed, after the assumption of the typical egg form, from the fragments scattered through the protoplasm.
8. Segmentation is very irregular and nuclear division often outruns the segmentation of the egg.
9. The ectoderm is formed by radial delamination of the two outer layers of cells.
10. The embryo escapes as an actinula with both basal and buccal tentacles.

The results obtained in this investigation differ in several points from those of Agassiz, whose description of *Parypha crocea* is the only one that I have found. Clark ('93), to be sure, refers to the eggs and spermatozoa of this species, but gives no account of them. Agassiz states that he was unable to

find any trace of eggs, and that the embryos are developed from a large spherical portion which buds off from a granular mass of protoplasm formed by the separation of the endoderm and ectoderm in the medusoid bud. This granular mass he calls the "germ basis." A study of stained specimens in section shows clearly that this granular mass, or "germ basis," as he calls it, is really the mass of sex cells which have already been described. His opinion that the embryo was formed by the budding off of large portions of this mass probably arose from the fact that in the early stages of the development the eggs are packed closely together and the membranes are indistinct, so that the whole mass appears somewhat homogeneous. As the eggs grow, they become less granular and in time are entirely separated from the germ tissue. As to the radial canals, they would probably be overlooked, except in sections, as they are never functional. The tentacles of the embryo are, however, so well developed that it seems strange that he should not have observed them, since he has noted tentacles upon the female gonophore where they are less clearly defined.

SYRACUSE UNIVERSITY, May, 1900.

BIBLIOGRAPHY.

- 1862. AGASSIZ, L. *Contributions to Nat. Hist. of U. S.* Vol. iv.
- 1865. AGASSIZ, A. *American Acalphae.*
- 1871. ALLMAN. *Monograph of Gymnoblastic Hydroids.*
- 1877. ALLMAN. *Report on Hydroids. U. S. Coast Survey.*
- 1880. BALFOUR. *Comparative Anatomy.*
- 1883. BOURNE. *Recent Researches upon Origin of Sex Cells in Hydroids. Quart. Journ. Micr. Sci.* Vol. xxiii, p. 617.
- 1892. BRAEM. *Origin and Development of Reproductive Cells in Tubularia. Journ. R. Micr. Soc.* p. 50.
- 1894. BRAEM. *Ueber die Knospung bei mehrschichtigen Tieren, insbesondere bei Hydroiden. Biol. Centralbl.* Bd. xiv, p. 140.
- 1888. BROOKS. *A New Method of Multiplication in Hydroids. Journ. R. Micr. Soc.* p. 433.
- 1894. BUNTING. *Origin of Sex Cells in Hydractinia and Podocoryne. Journ. of Morph.* Vol. ix, p. 203.

1897. CHUN. Histologie bei Hydromedusen. Leipzig.
1888. CLARK, SAMUEL F. Hydrozoa. *Riverside Nat. Hist.* Vol. i, p. 80.
1896. DOFLEIN. Die Eibildung bei Tubularia. *Zeitschr. f. wiss. Zool.* Bd. lxii, p. 1.
1892. GERD. Zur Frage über die Keimblätterbildung bei den Hydromedusen. *Zool. Anzeiger.* Bd. xv, p. 312.
1889. HARGITT. Preliminary Report on Reproductive Elements of Eudendrium. *Proc. Amer. Assoc. for Adv. of Sci.*
1900. HARGITT. Natural History and Development of Pennaria. *Amer. Nat.* Vol. xxxiv.
1884. HARTLAUB. Beobachtungen über die Entstehung der Sexualzellen bei Obelia. Leipzig.
1890. HICKSON. Development of Allopore. *Quart. Journ. Micr. Sci.* Vol. xxx, p. 579.
1891. HICKSON. Medusae of Millepora murrayi and Gonophores of Distichopora and Allopore. *Quart. Journ. Micr. Sci.* Vol. xxxii, p. 375.
1893. HICKSON. Development of Distichopora violacea. *Quart. Journ. Micr. Sci.* Vol. xxxv, p. 129.
1887. ISHIKAWA. Origin of Male Generative Cells in Eudendrium racemosum. *Journ. R. Micr. Soc.* p. 968.
1881. KLEINBERG. Development of Ova in Eudendrium. *Journ. R. Micr. Soc.* p. 256.
1892. LANG. Budding in Hydroids. *Amer. Nat.* Vol. xxiv, p. 1043.
1894. LANG. Zur Frage der Knospung der Hydroiden. *Biol. Centralbl.* p. 682.
- LANKESTER. Hydrozoa. *Enc. Brit.* Vol. xii.
1894. McMURRICH. Invertebrate Zoölogy.
1886. METSCHNIKOFF. Embryologische Studien an Medusen. Wien.
1899. SMALLWOOD. Pennaria tiarella. *Amer. Nat.* Vol. xxxiii.
1882. VARENNE, DE. Recherches sur les polypes Hydraires.
1881. WEISMANN. Ursprung der Geschlechtzellen bei den Hydroiden. *Zool. Anzeiger.* Bd. iii, pp. 226, 567.
1880. WEISMANN. Die Entstehung der Eizellen in der Gattung Eudendrium. *Zool. Anzeiger.* Bd. iv, p. 111.
1883. WEISMANN. Die Entstehung der Sexualzellen bei den Hydromedusen. (Monograph.)
1885. WEISMANN. Die Entstehung der Sexualzellen bei den Hydromedusen. *Biol. Centralbl.* Bd. iv, p. 33.
1892. WILSON. Variation in Yolk-Cleavage of Renilla. *Zool. Anzeiger.* Bd. xv, p. 545.
1896. WILSON. The Cell.

EXPLANATION OF PLATE I.

FIGS. 1-7 $\times$ 190; FIG. 8 $\times$ 270.

FIG. 1. Young medusa bud showing the formation of the germinal cells from ectoderm of bud. *ec*, ectoderm; *en*, endoderm; *h*, germinal cells.

FIG. 2. Later stage, germinal cells separated from the ectoderm of the bud.

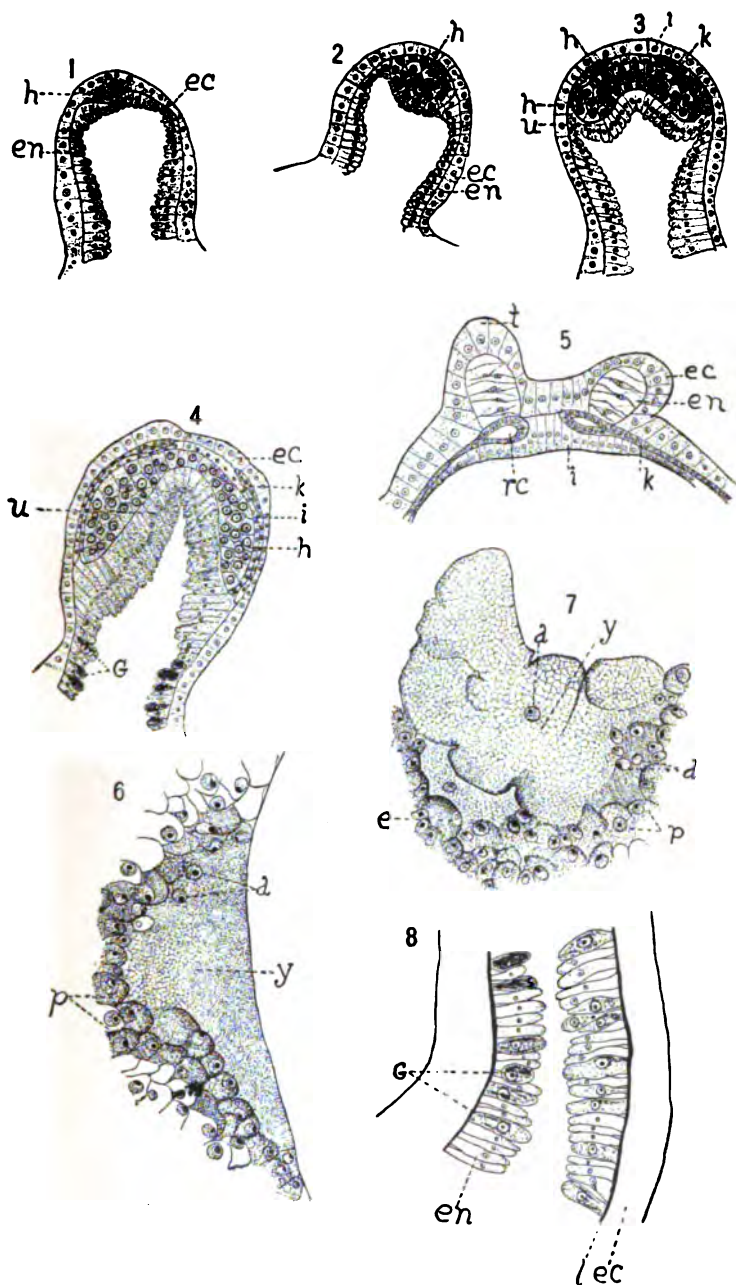
FIG. 3. Still later stage showing the mode of formation of the endodermal layer of the bell (*h*), the inner ectodermal layer (*i*), and the manubrium (*m*).

FIG. 4. Showing the layers of the bell completely formed.

FIG. 5. Distal end of mature female gonophore showing tentacles (*t*), rudimentary radial canals (*rc*), outer ectodermal layer of the bell (*ec*), and endodermal layer (*h*), inner ectodermal layer (*i*).

FIGS. 6, 7. Showing growth of the egg by the absorption of the primitive egg cells (*p*). Syncytium thus formed (*y*); nuclei of absorbed cells (*a*); young growing cell (*e*).

FIG. 8. Longitudinal section through peduncle from female head showing gland cells (*G*).



EXPLANATION OF PLATE II.

FIG. 1 $\times 127$; FIG. 2 $\times 118$; FIGS. 3-8, 10-15 $\times 765$; FIG. 9 $\times 495$.

FIG. 1. Growth of ovum (*e*) by absorption of primitive cells (*p*).

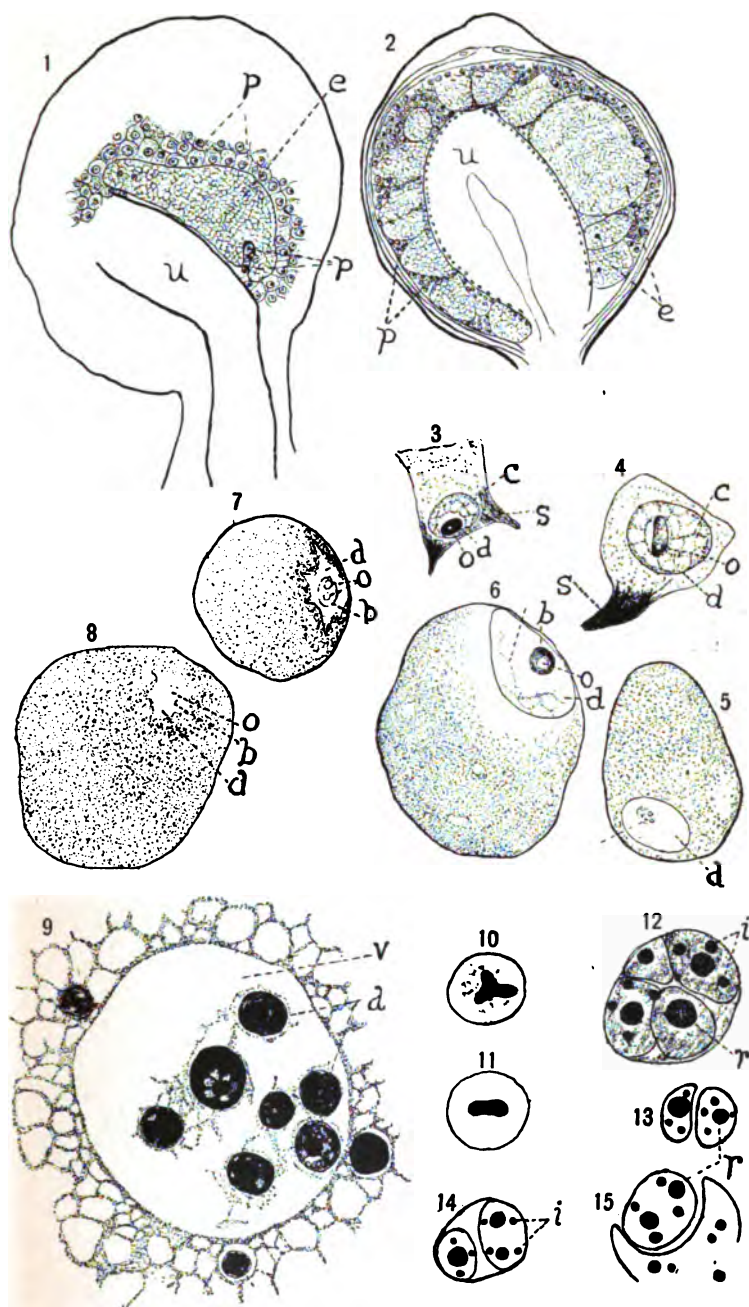
FIG. 2. Showing the number of growing primitive egg cells to be found in a single gonophore.

FIGS. 3, 4. Primitive egg cells in early stage of development showing pseudopodia (*s*); nucleus (*d*); oil drops (*o*); chromatin fibers (*c*).

FIGS. 5-8. Later stages showing the disappearance of the nucleus of the growing egg. *d*, nucleus; *b*, nucleolus; *o*, oil drops.

FIG. 9. Vacuole (*v*) in segmenting egg showing nuclei of absorbed cells (*a*) in various stages of disintegration.

FIGS. 10-15. Retrograding nuclei of absorbed cells. Nucleolus (*r*); assembled chromatin fibers (*s*). In Fig. 15 a portion of the nucleus has been absorbed; the smaller part is in the process of division, the nucleolus having already divided.



EXPLANATION OF PLATE III.

FIGS. 2, 6 $\times$ 115; FIGS. 1, 3-5, 7 $\times$ 152.

FIG. 1. Young ovum showing three nuclei (n). n^t in the process of division. No segmentation planes visible; a , nuclei of absorbed cells.

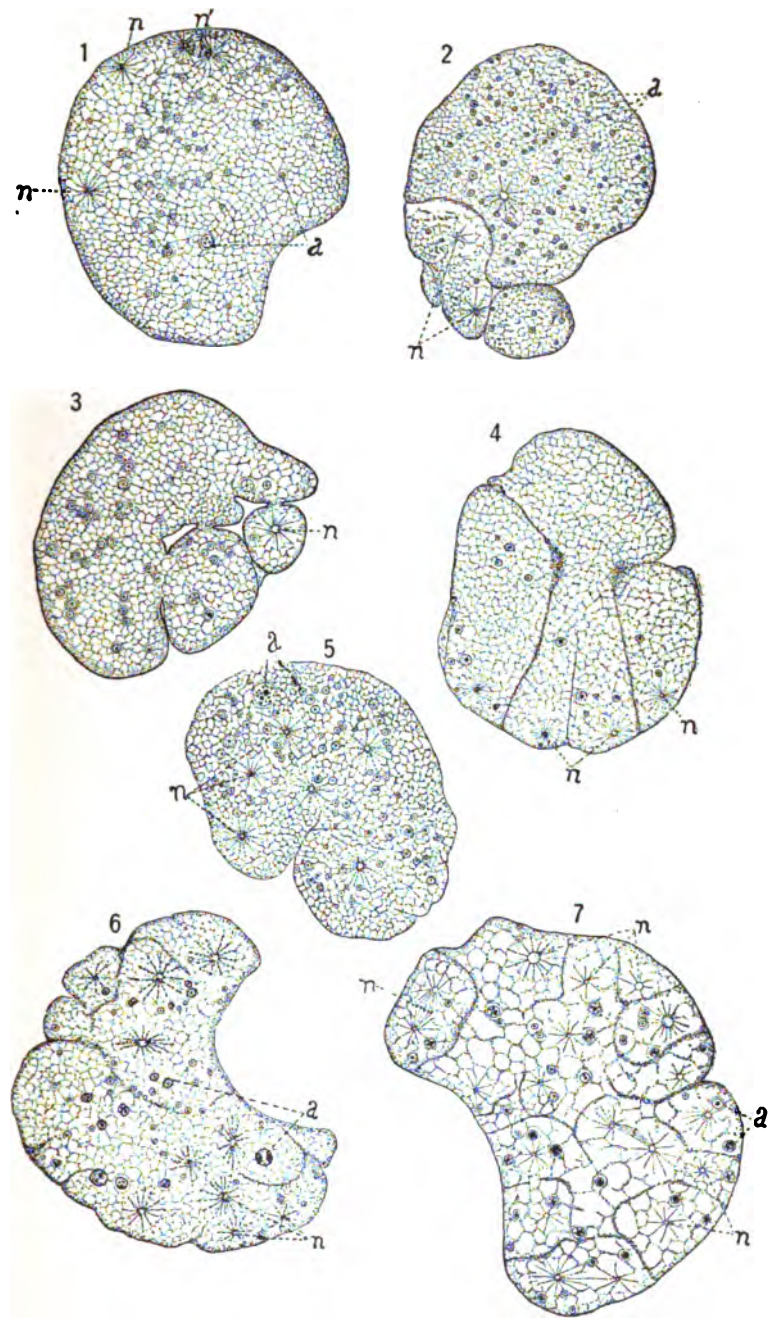
FIGS. 2, 3. Slightly later stages; segmentation planes well marked. n , nuclei; a , nuclei of absorbed cells.

FIG. 4. Still later stage of segmentation.

FIG. 5. Section through an egg containing fifteen or sixteen nuclei, but no well-defined cell walls. n , nuclei; a , nuclei of absorbed cells.

FIG. 6. Later stage; cell boundaries indefinite.

FIG. 7. Advanced stage of segmentation; cells irregular in outline, cytoplasm very reticular; n , nucleus; n^t , nuclei in process of division; a , nuclei of absorbed cells.



BIOLOGICAL BULLETIN.

A STUDY OF SOME TEXAN PONERINAE.¹

WILLIAM MORTON WHEELER.

THE observations hitherto published on the habits of the Ponerine subfamily of ants are extremely meager. This gap in our knowledge of ant life must be the more keenly felt by all myrmecologists because the Ponerinae (including the Cera-pachyi, a group allied to the Dorylinae, or driver ants) are, to all appearances at least, the most generalized of existing Formicidae. That the Ponerinae have a very primitive morphological and social organization seems to be indicated by the facts, first, that their colonies are composed of a relatively small number of individuals — like the incipient colonies of the more specialized Myrmicine and Formicine ants, and, second, that the polymorphism of the female is still, apparently, in an unstable condition, *i.e.*, the workers often differ little if any from the queens, or females, in form and size, and the two phases are frequently connected by individuals of an intermediate character (ergatoid, or ergatomorphic females). It has also been suspected by Professor Forel and Professor Emery that the breeding habits of the Ponerinae would be found to be of a primitive nature.

The common occurrence of three beautiful species of Ponerinae in the environs of Austin, Texas, has induced me to undertake the following study of their habits. The ants were observed in a state of nature or kept in jars of earth or in

¹ *Contributions from the Zoölogical Laboratory of the University of Texas.* Director, W. M. Wheeler. No. 6.

artificial nests of the Lubbock and of the Janet pattern. In collecting material for these nests, I have been considerably aided by two of my students, Mr. A. L. Melander and Mr. C. T. Brues. Mr. Brues also aided me in drawing some of the figures

that accompany this paper.

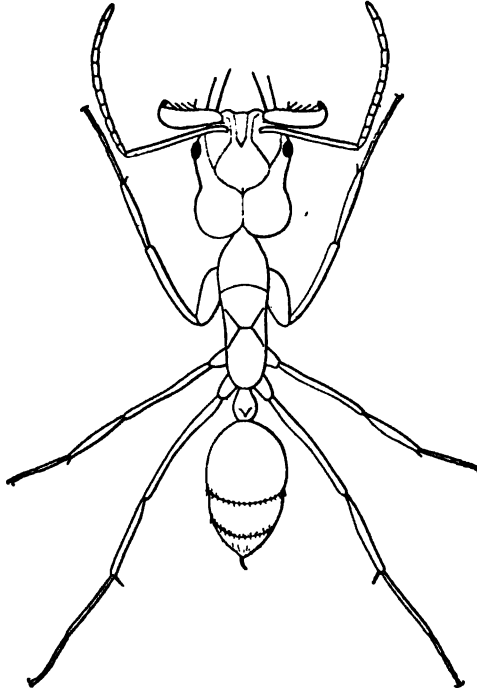


FIG. 1.—*Odontomachus haematodes* Linn. Worker.

The three species of Ponerinae which I have studied are: *Odontomachus haematodes* L.; *Pachycondyla harpax* Fabr. (= *Ponera amplinoda* Buckley); and *Leptogenys elongata* Buckley.¹ To European myrmecologists both *O. haematodes* and *P. harpax* are well-known species. They have a wide geographical distribution, occurring from Texas through Mexico, Central America and Brazil to Bolivia and Paraguay. *O. haematodes*

is also recorded from Georgia, Florida, and the Antilles.² In the vicinity of Austin, *P. harpax* and *L. elongata* are found under

¹ Specimens of the last species were kindly identified for me by Mr. Pergande as *Lobopelta elongata* Buck.? The query seems to be due to Buckley's wretched description of his *Ponera elongata*. Since *Lobopelta* is treated as a subgenus of *Leptogenys* by Forel, and since the species is very common in Texas, and therefore, in all probability, the one described by Buckley, I have changed the generic name and omitted the query. Should doubts arise, my figures (Fig. 4) will, I trust, enable any future observer to recognize the species.

² See Emery, "Beiträge zur Kenntniss der nordamerikanischen Ameisenfauna," *Zool. Jahrb.*, Abth. f. Systematik., Bd. viii, pp. 268, 269; Patton, *Amer. Nat.*, July, 1894, pp. 618, 619; Forel, *Biologia Centrali-Americana*, Hymenoptera, vol. iii, April, 1899, p. 21.

stones or logs, on moderately dry hill-slopes, often in the shade of trees or bushes, more rarely in the open fields. In these same localities *O. haematodes* does not occur. The few nests which I have seen belonging to this species were under stones lying on a dry sandy loam in a different part of Travis County. The exact distribution of these forms must be left for future determination.

It is not an easy matter to ascertain the exact number of individuals in any colony of the three species of Ponerinae. When the stone or log that serves as a roof to the nest is lifted, some of the ants always manage to escape, either into the surrounding vegetation or into their burrows. This is especially true of *L. elongata* and *P. harpax*, both of which, when excited, are very rapid in their movements. As a rough estimate it may be said that the nests of *L. elongata* contain from 10 to 50, those of *P. harpax* from 15 to 100, of *O. haematodes* from 100 to 200 individuals.¹

The nests of the three Ponerinae agree in being of a very primitive structure. They consist of a few simple and irregular burrows, or galleries, some of which run along the surface of the soil immediately beneath the stone or log, while others extend down into the soil obliquely or vertically to a depth of 8 or 10 inches. These burrows may anastomose, but they are not widened at certain points to form chambers, as in the nests of the more specialized ants (*Atta*, *Pogonomyrmex*, *Camponotus*, etc.). Even in artificial nests of the Lubbock pattern the Ponerinae dig only anastomosing galleries scarcely more than a centimeter in diameter.

Owing to the interest which attaches to the sexual phases of the Ponerinae, considerable effort was made to secure both the winged and apterous forms of the Texan species. Of *O. haematodes* only workers were seen, but as the full series of phases of this species—including the winged male, winged female, and the apterous ergatoid female, as well as the worker

¹ In tropical America (*teste* Forel, *loc. cit.*, *passim*) *P. harpax*, race *Montesumia*, Smith, *O. haematodes*, and the species of *Leptogenys* make their nests also in rotten wood, and the first species is said to nest also in dry, hollow stems. The colonies of *O. haematodes* are probably more numerous in individuals in the tropics than at Austin.

—have been made known through the labors of European myrmecologists, I fixed my attention on the other species. I append a brief description of these species before recording their habits.

P. harpax (Figs. 2 and 3) is a large, rather robust, subopaque, black ant, which carries its body close to the ground. It has a peculiar habit of folding its antennae and of peeping out of holes and crevices in the soil like a rat. In nearly all the nests

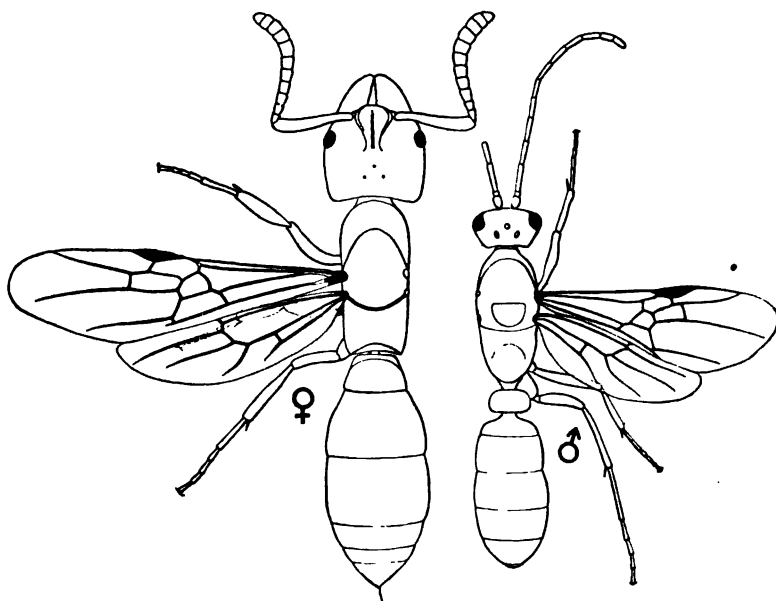


FIG. 2. — *Pachycondyla harpax* Fabr. Winged male and female.

that were opened only wingless individuals like the one represented in Fig. 3 were found. Except for a slight variation in size they were all alike externally, and I at first regarded them all as workers; but on dissecting several of these ants during the breeding season (from the 1st of March to the end of May) some of them were found to contain mature ova, while others had abortive ovaries. In a nest consisting of twenty-nine individuals captured May 27, dissection revealed the presence of mature eggs in thirteen, and one-third to one-half grown eggs in four of the ants. The ants were also caring for several

packets of eggs which they had laid. Like the pigeons among birds, *P. harpax* lays only two eggs at a time. I infer this from the fact that only two eggs are found to be mature in the ovaries. Since, in addition to these facts, wingless individuals actually laid eggs in my artificial nests, I feel certain that forms

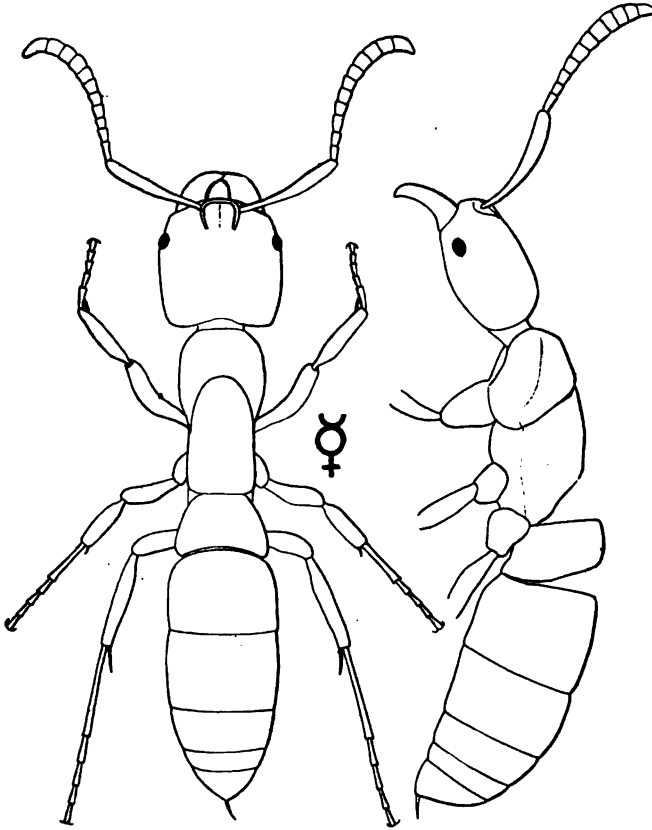


FIG. 3. — *Pachycondyla harpax* Fabr. Worker, dorsal and lateral aspect.

externally indistinguishable from the workers commonly function as females. These are, therefore, ergatoid or ergatomorphic females. There is not even a slight difference in size to separate the two forms externally, since some of the ergatoid females are smaller than some of the workers. There are, however, in certain nests of *P. harpax* winged females of a less

worker-like aspect. These were seen only twice — once in a large nest found under a log on March 12, and again in a nest discovered in a very different locality by Mr. Brues, March 31. In both nests there were many of these winged females resembling Fig. 2, ♀. They had small but distinct ocelli. Several of them had lost some of their wings, while a slight touch caused others to lose these appendages. Before finding these winged females, I had found the winged males as early as March 3 in a small nest containing ergatoid females and workers. There were two of these males clinging to the under surface of the stone that covered the surface galleries of the nest. They differ so much from the apterous females and workers, that, if taken on the wing, they would scarcely be regarded as ants (*cf.* Fig. 2, ♂, and Fig. 3). They are much shorter and more slender than the workers, and their long, slender, 13-jointed antennae are not geniculate, but straight, with the basal and second joints short and the remaining joints slender and subequal. The eyes and ocelli are very large and prominent. These and other equally pronounced structural peculiarities in the thorax and abdomen are shown in Fig. 2, ♂. The small mouth-parts, the hypopygium, trochanters, tibiae, and tarsi are honey-yellow; the venter and sides of the abdomen are piceous.¹ On the 31st of March I found four nests containing winged males. Three of these nests each contained three males, the remaining nest contained only two.

L. elongata is smaller and far more slender than *P. harpax*, with long delicate legs and antennae. The specific name is very applicable not only to the adult ant but also to the egg, larva, and pupa. The worker (Fig. 4, ♀) is of a rich claret red color, becoming yellow towards the tip of the abdomen. In form, color, and movement this ant is certainly the embodiment of elegance. The workers which were dissected — some thirty specimens from different nests — contained only minute and abortive ova, and for some time I failed to find the queen.

¹ To my knowledge no description has hitherto been published of the male of the typical *P. harpax*. I infer, however, from Forel's contribution (*loc. cit.*, p. 12) that Smith has described (*Cat. Hym.*, vol. vi, p. 108, 1858) the winged male of his *P. montezumia*, a form which Forel regards as merely a Mexican race of *P. harpax*.

Finally I noticed in each nest a single ant with a somewhat larger abdomen than that of the workers, but, like them, without any traces of wings. Dissection of three such individuals disclosed mature eggs — only a single pair in each insect, as in *Pachycondyla*. There could be no doubt that I had found the hitherto unknown female of *Leptogenys*.¹ They have no ocelli,

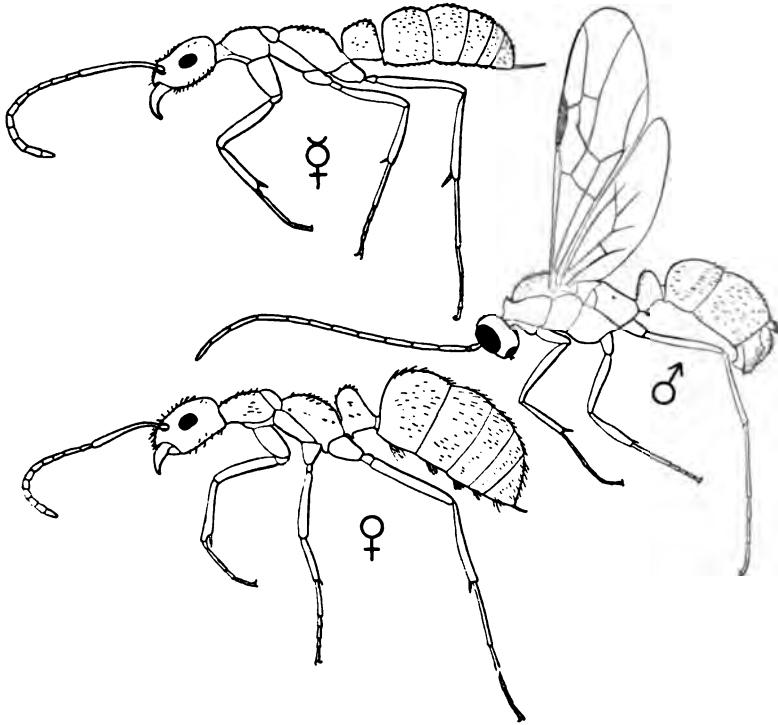


FIG. 4. — *Leptogenys (Lobopelta) elongata* Buck. Worker, winged male, and apterous female.

but the shape of the node and of the abdomen serves to distinguish them at once from the workers (see Fig. 4, ♀). They are certainly ergatoid females, but as such they differ from the ergatoid females of *P. harpax* in several respects: First, they resemble the workers less closely; second, they occur singly in the nests; and, third, they have the status of true queens,

¹ Thus Emery's supposition (Emery, C., "Zur Biologie der Ameisen," *Biol. Centralbl.*, Bd. xi, p. 174, 1891), that the females of this genus are apterous, is proved to be true of at least one species.

because they are the only known egg-laying individuals of the species. The winged, light-yellow male of *L. elongata* was determined on circumstantial evidence, as it could not be found in the natural nests, nor bred from the cocoons in the artificial nests. What seemed to be the males (Fig. 4, ♂) were found during the autumn and spring flying about the lights in the houses. They are somewhat smaller than the workers, have very large eyes and ocelli, and the node is shaped like that of the female. When placed in the artificial nests of *L. elongata* they are not only not molested, but move about among the workers as if they had always belonged among them. To one who has witnessed the hostility of these ants to any ant of a different species placed in their nest, this amicable reception amounts to a demonstration of specific identity.

During the course of my observations I have had frequent opportunity to observe the conduct of all three species of Ponerinae towards ants of the same or of different species introduced into their nests. They are always hostile to ants of a different species, but the aggressive nature of this hostility varies according to circumstances. If one or a few strange ants be placed in a nest of Ponerinae, they will be at once attacked and killed by concerted action, but if a single Ponerine be placed in the nest of another species — say in a nest of the agricultural ant (*Pogonomyrmex barbatus*) — it will if attacked defend itself valiantly with its mandibles and sting, but it will prefer to seek refuge in some corner of the nest and remain concealed. Single specimens of *P. harpax*, introduced at different times into the nest of *P. barbatus*, managed to evade the pugnacious agriculturals and to remain alive for days. Whenever they left their concealment, however, they would be attacked by a swarm of agriculturals, but the Ponerinae were so supple and fierce that they always managed to throw off their assailants and to conceal themselves. Other investigators, like Forel and Wasmann, have called attention to this difference in the pugnacity of ants according to whether they find themselves alone or backed by numbers of their own species.

The species of Ponerinae differ in their reception of ants of the same species from other nests. If two nests of workers

of *P. harpax* (including ergatoid females) be thrown together into the same jar, the ants of different nests at once begin to struggle with one another. One ant will grasp a stranger by the head with her mandibles and smite her with rapidly vibrating antennae; or the two foes will interlock mandibles and pull in opposite directions till one gives up and runs away, only to begin the same performance with some other stranger. This peculiar tugging goes on between the ants of different nests for hours, or even for three or four days. Then, without any mutilations or deaths as a result of the struggle, the ants abandon all this hostile play, and thenceforth form a single peaceable community.

When two nests of workers of *L. elongata* are put together, the struggle which ensues presents a slightly different aspect. An ant from one nest will grasp a stranger by an antenna or by one of her fore feet and, while tugging, smite her opponent with very rapidly vibrating antennae. In this species there is no interlocking of mandibles. After several days of tugging and struggling the same peaceable conditions supervene as in the case of *P. harpax*.

Workers of *O. haematodes*, belonging to different nests, never display the slightest animosity towards each other, so far as I have been able to observe: in a strange nest they are treated exactly as if they had always belonged there. An exception to this conduct of *O. haematodes* in the case of friends deprived of their antennae will be mentioned below.

A few observations were made on the conduct of nests of workers towards sexual individuals from other nests. One of the two males of *P. harpax*, taken March 3, was placed in a strange nest of ten workers (including ergatoid females). The male, after walking about on the earth for a short time, found his way into one of the galleries in which six of the wingless individuals were digging. On perceiving the male they at once stopped their work and began licking the winged stranger with signs of great agitation and affection. At the same time they carefully massaged all his limbs and segments with their mandibles. During this effusion the attitude of the male was most peculiar. He lay on his back in what may be described

as a pupal attitude, with his legs drawn up against his body, and his long filiform antennae folded against the mid-ventral surface of his thorax and abdomen. This amusing scene was still in progress when I had to quit my observations half an hour later. The next morning I saw one of the workers (or an ergatoid female?) carry the male in her mandibles a distance of four inches and disappear with him into one of the galleries. During transport the male preserved the motionless pupal attitude. This treatment of a strange male contrasts strikingly with the treatment received by the male which was allowed to remain with the workers and ergatoids of his own colony. After eight days of captivity this male was killed and devoured by one of the wingless individuals. When I first observed this Amazon, she had already consumed the small head of her victim and was hanging from the roof of a gallery, twirling the torso about with her feet and rasping away the thoracic muscles with her maxillae. Was this act prompted by hunger? or had the male fulfilled his mission in this nest already provided with several packets of eggs? I have already mentioned the fact that the male of *L. elongata* was received by a strange colony of workers like an old acquaintance. The absence of any display of affection in this case may have been due to the fact that there were no females in the nest. Two strange females introduced into this same nest at different times were at once surrounded and attacked by the workers, and although the colony was without a queen, they were either maimed or killed during the course of the day.

Many of the habits of the Ponerinae closely resemble those of other ants. All the habits relating to the cleanliness of the individual ants and of the nests are the same as those of the Myrmicinae and Formicinae. They clean themselves and one another with great care. The males spend much of their time drawing their delicate antennae through the strigils on the fore feet. The nests of all the species are kept scrupulously clean — all refuse, inedible fragments of their insect prey, empty cocoons, dead sister ants, etc., are carefully deposited in one of the corners of the nest exposed to the light and as far as possible from the chambers in which the young are reared.

In digging a gallery *P. harpax* first uses its feet and powerful mandibles in removing the earth, after the manner of the digger-wasps, but as soon as the gallery is sunk to a certain depth, only the jaws are used in removing the lumps of earth.

Emery's observations¹ show that the Ponerinae have the power of stridulating. He has even been able to produce the sound in preserved specimens of certain American species, including species of *Pachycondyla*. Although the ants observed by me occasionally moved their abdomens as if stridulating, no sound could be detected.

The sting of the three species of Ponerinae, although of formidable length, does not inflict severe pain, at least when compared with the sting of the agricultural ant (*Pogonomyrmex barbatus* Sm.). The pain may be acute for ten or fifteen minutes, but then disappears, often without even reddening the skin; whereas the sting of *Pogonomyrmex* produces a throbbing pain which endures for hours and is sometimes accompanied by a sensation of sickness.

The Ponerinae do not seem to feed one another, like the specialized ants. In captivity *P. harpax* would eat the yolk of egg or even sugar, but it would not eat termites. *L. elongata* devoured termites and small caterpillars with avidity, but would not eat flies. *O. haematodes* is more omnivorous; besides caterpillars, house-flies, beetles, and small Hemiptera, it will eat sugar, bread, cake, etc.

Pachycondyla and *Leptogenys* are like other ants in their methods of killing their prey or in feeding upon it, but *Odonotomachus* is most exceptional in these particulars. Its whole life, apart from the care of its young, appears to center in its peculiarly constructed mouth. The long linear mandibles with hook-shaped dentate tips are inserted close together. They are usually carried wide open, as represented in Fig. 1, while the ant is moving about in search of food or while it is feeding. The cutting edges of the mandibles are furnished with some sensory hairs, two of which, nearly as long as the mandibles, are inserted near the base and point directly forward

¹ Emery, C., "Zirpende und springende Ameisen," *Biol. Centralbl.* Bd. xiii, pp. 189-190. 1893.

when the mouth-parts are in the position represented in Fig. 1. The slender antennae, too, are carried in a peculiar position, their tips being directed inwards. The touching of a living insect or of any unfamiliar object with these incurved tips calls forth a peculiar response, which seems to be largely of a reflex nature. The ant darts forward and suddenly closes its mandibles with a very audible click. The signal for closing the mandibles seems to be given the moment the long sense-hairs touch the object. The mandibles are brought together with such force that if they strike a solid object the ant is thrown backwards — often to a distance of three or four inches — occasionally even to a distance of ten or twelve inches. The ant alights on its feet, like a cat, and again advances to repeat the act. This remarkable clicking and leaping habit is called into play on every occasion, and its study discloses some interesting facts, as the following jottings from my notebook will show :

May 12. Placed a living house-fly in the *Odontomachus* nest. Its movements at once attracted several ants, which began snapping at it like a pack of angry dogs. With each snap a leg or wing was severed and often thrown to a distance of 2 or 3 inches. In less than a minute all the limbs had been shorn from the trunk. The fly was then seized and decapitated. Next the following living ants were placed in the nest in succession: *Eciton sumichrasti*, *Pogonomyrmex barbatus*, and *Leptogenys elongata*. The first and last were reduced to torsos as rapidly as the fly; the harder legs and antennae of the *Pogonomyrmex* offered greater resistance, but not for long. The stings and strong mandibles of these various ants were no protection against the singular method of attack adopted by the *Odontomachus*. A smooth green caterpillar was next introduced into the nest. It was at once surrounded and attacked by a dozen ants. At first the ants retreated from the writhing larva after each snapping of the mandibles, but soon they grew bolder and, retaining their hold, drove their stings through its velvety skin. One ant with a dexterous movement closed its mandibles in the caterpillar's brain. In a few moments the green blood of the victim was oozing from a score of wounds.

May 25. *O. haematodes* reacts to lifeless objects just as it does towards living insects. If the points of a pair of tweezers be held in the nest and rapidly opened and closed, they at once become the center of a swarm of snapping and leaping ants. A small glass vial dropped into the nest at once begins to tinkle under the blows of the alternately advancing and retreating emmets. A lump of sugar is attacked in the same manner for some moments, till the ants learn that it is edible, when they settle down on it and lap its crystals with avidity. Even soft viscid or liquid substances, like the yolk of egg, poured on the floor of the nest, at once release the same peculiar response. As soon as the ants perceive it with the contact-odor sense of their antennae, they begin snapping at it as if it were a deadly foe. In this case the tips of their mandibles are smeared with the yolk. This causes them to feel some discomfort apparently, and they wipe them on the hard floor of the nest, much as a bird would wipe its beak in similar circumstances. It is only after several repetitions of this performance that they approach the yolk without closing the mandibles. Then they begin to eat it, and some hours later they begin to cover it up with little pellets of earth, for *O. haematodes*, as well as *L. elongata* and *P. harpax*, share with the Myrmicine and Formicine (and I may add, also, the Doryline ants!) the habit of burying offensive substances.

Observers¹ have called attention to the fact that ants deprived of their antennae are in some cases attacked and killed by ants of the same nest. A few experiments were tried for the sake of testing these observations on *Odontomachus*. One of these ants, deprived of both antennae, was replaced in the nest from which it had just been taken. It was at once fiercely attacked by several ants as if it had been a strange insect. It stopped, as if dazed, unable to meet its sister workers with the customary antennal greeting. The ants, however, soon perceived their error, and a few hours later one of them was licking and fondling the mutilated ant. By the next day the mutilated ant was dead, probably as a result of the

¹ Forel, Fourmis de la Suisse, p. 119, *et al.*; Wasmann, Die zusammengesetzten Nester und gemischten Kolonien der Ameisen, pp. 151, 152, Münster i. W., 1891.

extirpation of its antennae. Two other experiments terminated in the same way.

Two inferences may, I believe, be drawn from these observations on the snapping and jumping habits of *Odontomachus*: First, it seems evident that these are reflex actions; second, that they can be inhibited by the will of the insect. This latter conclusion is supported by the experiment with the sugar and the yolk. It also shows that these ants learn by experience, and that they possess some form of memory.

The peculiar snapping and leaping habits of *O. haematodes* have been remarked by a few other observers. Emery has published a brief note on this subject,¹ and Forel² says of this insect that "en Colombie on l'appelle 'Fourmi tac', à cause du bruit qu'il fait en refermant brusquement ses mandibules. Par le même mouvement il ressaute en arrière lorsqu'il les referme contre un objet, ce qui lui a fait attribuer à tort la propriété de sauter." I cannot accept this last statement, which seems to be a contradiction in terms. The ant makes use of its saltatory powers for purposes of escape, as any one who tries to capture a colony of these ants may readily observe. That it leaps backward instead of forward, like other insects, is due to its using a most unusual organ for leaping, for the mandibles, which, as in other ants, are used for digging and transporting the soil, carrying eggs and larvae, and for killing and cutting up prey, have acquired an additional function as saltatory organs in *O. haematodes*.

The breeding habits and the characteristics of the eggs and larvae of the Ponerinae exhibit striking deviations from those of other ants. I have not seen the eggs of *Odontomachus*, but throughout the month of May I have often happened on the eggs of *Pachycondyla* and *Leptogenys*. These are white and of a slender, oblong shape (Fig. 8, *a*), somewhat smaller in the latter than in the former genus. They differ in shape from the eggs of species of *Eciton*, *Camponotus*, *Formica*, *Pogonomyrmex*, *Solenopsis*, and *Tapinoma*; for the ants of these genera, representing several subfamilies, agree in having elliptical and

¹ *Biol. Centralbl.* Bd. xiii, pp. 189-190. 1893.

² *Loc. cit.*, p. 20.

much less slender eggs than the Ponerinae. The Ponerinae also keep their eggs in more regular packets, the long axes of the different eggs being placed parallel with one another. I have not been able to determine the length of embryonic development with accuracy. It seems to be much protracted, as in other ants. Some eggs of *P. harpax* in an artificial nest had not hatched at the end of five weeks.

The larvae of the Ponerinae differ from those of all other ants in several particulars, first made known by Emery.¹ Emery describes and figures the larvae of *Ponera stigma* Fab. (New Guinea), *P. caffraria* F. Sm. (Cape Colony), *Diacamma rugosum-geometricum* F. Sm. (Celebes), and *Odontomachus haematodes* L. (Cayenne). All of these larvae agree in having the mandibles powerfully developed for ant-larvae, the anterior portion of the body long and slender and folded over the abdominal portion, and in being covered with rows of peculiar tubercles beset with more or less prominent bristles.

The larvae of *Pachycondyla* and *Leptogenys* are here figured for the first time. I have seen fit to figure also the *Odontomachus* larvae because my material was evidently in fresher condition than that depicted by Professor Emery. The larvae of the three genera may be arranged in a series beginning with *Odontomachus* and terminating with *Pachycondyla*.

The young larva of *O. haematodes* is represented in Fig. 5, *a*, which shows the arrangement and character of the bristly tubercles and the neck-like anterior portion, consisting of the head, the three thoracic and the first two abdominal segments. The remaining eight abdominal segments are much enlarged and flattened ventrally. The larva is kept on its back, and the neck-like anterior portion rests against the flattened ventral surface. The shape of the tubercles, each of which is tipped with a rigid bristle and encircled with bristles, is shown in Fig. 5, *b*. The larva was about to moult, so that the tubercle of the succeeding cuticle is seen shining through the old one. The adult larva is shown in Fig. 6, *a*. Compared with that of the young larva, its head is very small in proportion to the body.

¹ "Intorno alle Larve di Alcune Formiche," *Mem. della Accad. delle Scienze dell'Istituto di Bologna*, 7. Maggio, 1899, 2. Tavole.

This seems to be the universal rule in ant larvae. The head in dorsal view is represented enlarged in Fig. 6, *b*. The powerful dentate mandibles lie just below the outer edges of the bilobed labrum; still lower and projecting forward lies the labium, bearing on its tip the opening of the spinning gland (to be used in weaving the cocoon), and on either side two peg-shaped tactile (?) organs. Similar but somewhat larger organs are seen on the edges of the maxillae, which protrude on either side below the mandibles. The ten tracheal stigmata, begin-

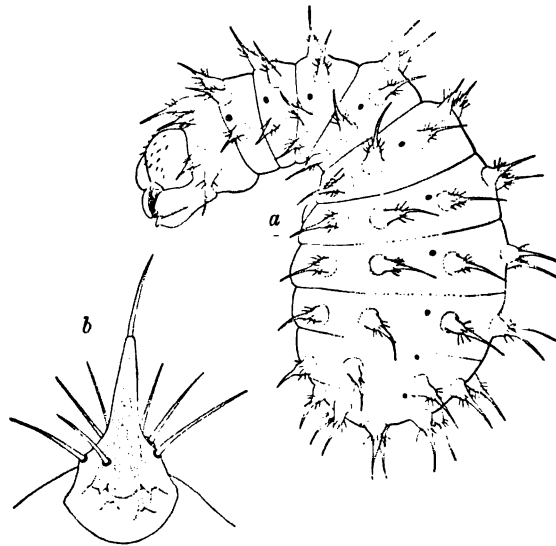


FIG. 5. — *Odontomachus haematodes* Linn. *a*, young larva; *b*, tubercle of the same.

ning on the mesothoracic and terminating on the eighth abdominal segment, are clearly shown in Fig. 6, *a*. The bristly tubercles are essentially the same in structure as those of the younger larva, but they are relatively shorter and smaller. (Cf. Fig. 5, *b*, and Fig. 6, *c*.)

The larvae of *Leptogenys* (Fig. 7) are remarkably slender and scarcely flattened on the ventral surface. In the young larvae (Fig. 7, *a*) the tubercles are distinctly curved and pointed, without apical bristle, and with only a few rather short bristles encircling the base (Fig. 7, *d*). In the adult larvae (Fig. 7, *b* and *e*) the tubercles are larger and shorter, with blunt or

acuminate apex and with relatively longer and more numerous basal bristles. The head of the adult larva (Fig. 7, *c*) is remarkable for its length and the narrowness of the labrum, which is nearly as long as the slender mandibles and provided with a median tooth at its tip.

The larvae of *Pachycondyla* (Fig. 8) are neither as slender as those of *Leptogenys* nor as robust as those of *Odontomachus*.

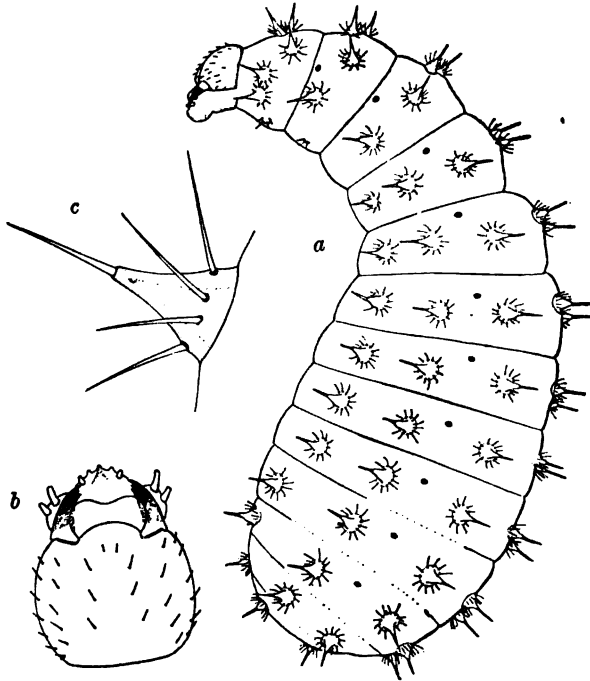


FIG. 6.—*Odontomachus haematodes* Linn. *a*, adult larva; *b*, head of same (dorsal aspect); *c*, tubercle.

The ventral surface of the abdomen is distinctly flattened. The head (Fig. 8, *e*) resembles that of *Odontomachus*, especially in the shape of the labrum and mouth-parts. There is a striking difference between the tubercles of the very young and the adult larva. In the former (Fig. 8, *b*, *c*) the tubercles are nearly or quite straight, and somewhat longer and more pointed than those of *Leptogenys*. They lack the terminal bristle. The bristles about the base are somewhat irregular in their insertion.

In the adult larva (Fig. 8, *d*) the tubercles are reduced to large more or less flattened bosses, encircled with a regular row of numerous, rather long bristles. In the stages between those figured the gradual flattening of the juvenile spine-like tubercles can be traced through the successive moults.

In this series of larval forms, *Odontomachus* seems to represent the most primitive condition. Here both young and old larvae have pointed, bristle-tipped tubercles, and there is little

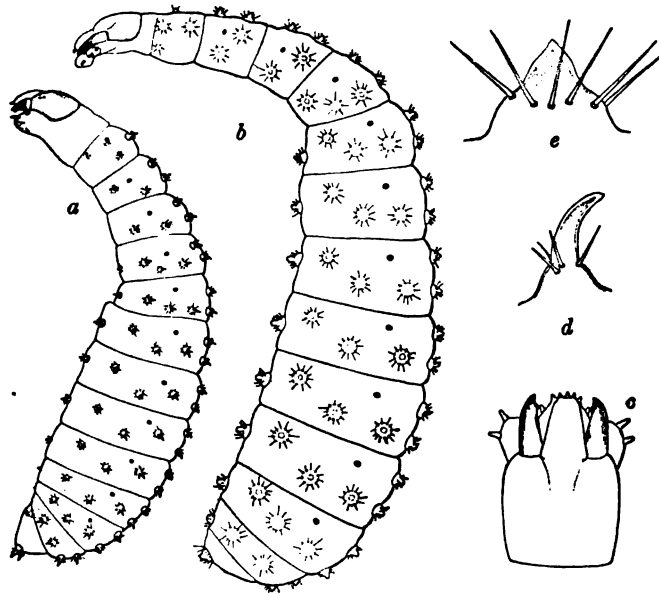


FIG. 7. — *Leptogenys (Lobopelta) elongata* Buck. *a*, young; *b*, adult larva; *c*, head of adult larva (dorsal aspect); *d*, tubercle of young; *e*, tubercle of adult larva.

difference between the tubercles of the young and adult. In *Leptogenys* and *Pachycondyla* the apical bristle is absent, but in both genera the young larvae have pointed tubercles. In the adult larva of the former genus there is a perceptible blunting of the tubercles, while in the adult larva of the latter the tubercles have nearly subsided.¹

¹ Emery's observations on *Ponera stigma*, *P. cafraria*, and *Diacamma geometricum*, seem to indicate conditions the reverse of those which I have described. Of the former species he says (*loc. cit.*, p. 4); "Nello stado piu giovane, si vedono solo deboli accenni dei tubercoli cutanei; ritengo che questo stado debba corrispondere alle larve di prima schiusa e che lo stado seguente, di poco piu grande, sia quello

The bristly tubercles of the larvae of the Ponerinae are so prominent as readily to suggest the question of their function. Prof. L. Biró, who made some observations on the larvae of *P. stigma*, which he sent to Professor Emery, believes that the pointed tubercles are organs of defense. He saw these larvae when disturbed by some termites move their long necks back and forth with sufficient force to drive away the intruders.¹

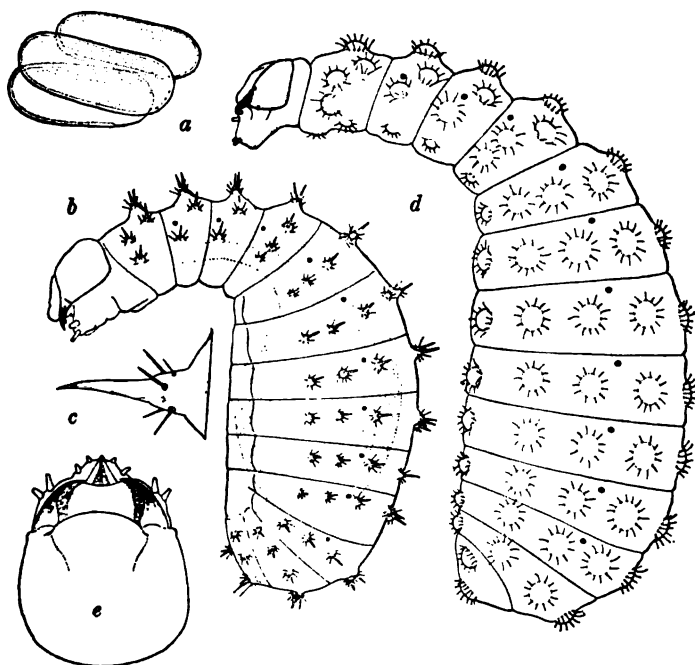


FIG. 8. — *Pachycondyla harpax* Fabr. a, eggs; b, young larva; c, tubercle of the same; d, adult larva; e, head of the same.

che segue la prima muta; questi si fanno successivamente piu numerosi e sporgenti, a misura che la larva cresce." In the larvae of *Diacamma* a very different condition is described: "Sopra ciascuno (segmento) di essi si trova un serie trasversale, irregolare di tubercoli conici, ineguali che, nelle larve piu sviluppate, portano da uno a quattro peli. Nelle piccole larve, i tubercoli sono piccoli, subcilindrici e senza peli; negli stadi intermedi passano per una forma acuminata con pochi peli."

¹ Professor Emery (*loc. cit.*, p. 4) quotes from Professor Biró's letter: "Nelle gallerie del nido scavato nel legno putrido, si trovavano le larve dal lungo collo, coperte di spini singolari: abbandonate dai loro vigliacchi custodi, quelle larve sapevano difendersi da se; quando qualche termite (il nido di queste trovavasi nello stesso legno) si avvicinava ad una di esse, questa batteva innanzi e indietro col suo collo di cigno e tosto veniva lasciata in pace."

Biró's observations may be true of *P. stigma* without being applicable to the three forms of Ponerinae which I have observed. In artificial nests I have seen the neck movements of the larvae, but they were often executed when the larva was undisturbed, except perhaps by the pangs of hunger, and they were not always made when termites or other insects were running about and over them. Moreover, we should expect to find the tubercles more highly developed on the neck than

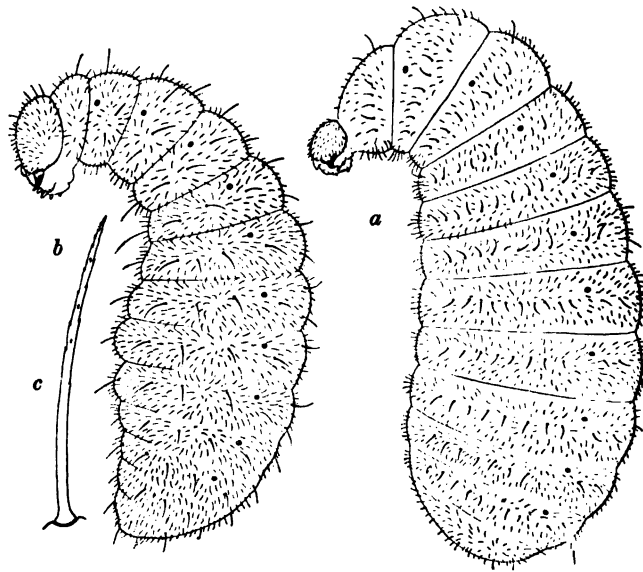


FIG. 9. — *Pogonomyrmex barbatus* Smith. *a*, nearly adult larva; *b*, young larva; *c*, serrated bristle of the same.

on the body, if they are really used as Biró suggests. I believe that while they may be organs of passive defense, like the somewhat similar tubercles and spines of certain caterpillars, they also fulfill other functions; they would seem to facilitate the carrying of the larvae either singly — when full-grown — or in batches — when young — by the worker ants. In the last instance they would represent a peculiar form of the “poils d'accrochage” carefully studied by Janet.¹ Janet finds that the young larvae of the more specialized ants are covered with

¹ Les Fourmis. Conférence faite le 28 Février, 1896. Paris, 1896.

hooked bristles, which cause them to adhere together in packets and thus facilitate their transportation by the workers. The appearance of these peculiar hairs in the young and half-grown larvae of one of our common Texas ants, *Solenopsis geminata* Fabr., is shown in Fig. 10. The very young larvae have only simple bifurcated hairs, but when half-grown they have on the dorsal surface of several of the segments, besides a much greater number of these simple bifurcated hairs, several rows of long and peculiarly contorted bristles, terminating in short

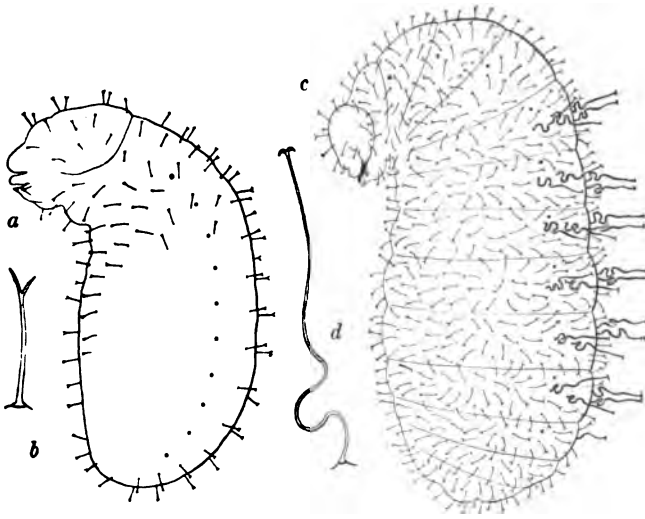


FIG. 10.—*Solenopsis geminata* Fabr. a, very young larva; b, furcate bristle of same; c, half-grown larva; d, contorted furcate bristle of same.

bifurcations. Still another modification of the “poils d'accrochages” is seen in *Pogonomyrmex barbatus* (Fig. 9), the young larvae of which have the longer bristles serrate on the apical half, so that they remind one of the hairs of certain mammals. All of these modifications—the bristly tubercles of the Ponerinae, the simple and contorted bifurcated bristles of *Solenopsis*, the serrate bristles of *Pogonomyrmex*, and possibly also the fascicles of uncinata hairs described and figured by Emery (*loc. cit.*) for the larva of *Sima natalensis* F. Sm.—seem to subserve the same purpose—a most interesting example of independent

lines of development terminating in organs of different structure but identical function.

The pupae of *Odontomachus*, *Leptogenys*, and *Pachycondyla* are enclosed in elliptical brown cocoons, like the pupae of many species of *Formica* and *Camponotus*. The pupa of *L. elongata* is remarkable on account of its very slender shape, a peculiarity not confined to the pupa, but, as we have seen, extending also to the egg, larva, and imago.

We come now to a consideration of the breeding habits of the *Ponerinae*. The little that has been made known concerning their habits has led European myrmecologists to believe that the philoprogenitive instincts of these ants must be less highly developed than those of the *Myrmicinae* and *Formicinae*. Thus, according to the above-quoted note of Professor Biró, when the nest of *Ponera stigma* is disturbed the ants flee and the larvae are "abbandonate dai loro vigliacchi custodi." And Professor Forel has made what appears to be a somewhat similar observation on our American *Ponera coarctata* Fabr. in North Carolina¹: "La *Ponera coarctata* américaine est très commune dans les troncs pourris et sous les pierres. J'ai fait chez elle une observation qu'il est bien difficile de faire en Europe; mais ici elle est tout à fait constante. Lorsqu'on découvre un nid de *Ponera* dans un tronc pourri, on voit leurs cocons jaunes assemblés dans un coin, mais absolument abandonnés des ♀ qui n'essaient pas de les sauver, ni de les recueillir. Par contre, elles prennent le plus grand soin des larves qu'elles emportent et cachent. Je soupçonne que chez ces fourmis, moins sociales que les autres, les nymphes sortent seules de leurs cocons, sans avoir besoin de l'aide des ♀."

These observations relate to species of *Ponera* and are at variance with the conclusions which I have reached from a study of three other genera of *Ponerinae*. In many of the nests which I have examined the total number of the eggs, larvae and pupae, could scarcely be greater than one and one-half to twice the number of the ants. This fact, together with what has been said of the small number of eggs laid at one time by a single female, shows very clearly that the *Ponerinae*

¹ *Ann. de la Soc. Entomol. de Belgique*. Tome 43, p. 443. 1899.

are not nearly so prolific as the species of *Camponotus*, *Formica*, *Pogonomyrmex*, *Pheidole*, *Tapinoma*, *Eciton*, etc. Indeed, the small number of ants in the nests of the Ponerinae is probably the direct result of this limited productivity. If this is the case, it does not seem probable that these ants would be more careless of their progeny than the very prolific specialized ants. On the contrary, we should expect them to extend even greater protection to their offspring. This my observations show to be the case; at any rate, *P. harpax*, *L. elongata*, and *O. haematodes* are in nowise inferior to the Myrmicinae and Formicinae in this respect. The slightest disturbance of the natural or artificial nests of these ants causes them at once to seize their eggs, larvae, and cocoons, and to make for their galleries. Occasionally some of the ants escape without anything, but if they are watched for a few moments, they will be seen returning, often in the very face of danger, to carry off more of their young. They are, it is true, most careful of their eggs, somewhat less careful of the larvae, and least careful of their cocoons; but these distinctions are not always apparent and can only be affirmed as the result of many observations. When the colony is agitated, it is probably most easy for the ants to seize and remove the small packets of eggs and the younger larvae, and least easy to carry off the larger larvae and the awkward cocoons. Dead pupae are often collected in one part of the nest and are there allowed to lie unheeded. I am inclined to think that Professor Forel may have seen such abandoned pupae in the nests of *P. coarctata*.

The strong development of the mandibles of the Ponerine larva as compared with those of other ants led Emery remotely to surmise the method which the Ponerinae employ in feeding their young.¹ But no myrmecologist could have predicted the

¹ *Loc. cit.*, pp. 8, 9. "Sembrami pertanto che lo sviluppo notevole della bocca e particolarmente delle mandibole, nelle larve delle Ponerinae e dell' *Acanthostichus* inducano a qualche supposizione relativamente alla biologia di queste formiche. Le larve delle specie europee che finora furono osservate vengono alimentate col contenuto dell' ingluvie delle operaie che queste regurgitano sulla bocca delle loro larve, e forse anche col secreto di ghiandole salivari. In queste specie, l'alimento delle larve consiste dunque esclusivamente di sostanze liquide o

remarkable and un-ant-like procedure which I have been able to observe in the three Texan species.

My first observation on this singular method of feeding the larvae was made on a large nest of *Pachycondyla* found under a stone at the foot of Mt. Bonnell, near Austin, May 5. Before the ants could carry them away, I had scooped up a fine lot of larvae, together with the earth in which they were lying. Among the larvae were several pieces, one or two segments long, of a recently killed myriopod (*Scutigera*). Into these pieces the larvae, some of which were nearly full-grown, had inserted their heads and were devouring the softer tissues! This could be distinctly seen with the pocket lens through the glass of the vial to which the larvae had been transferred. In another nest of the same species, uncovered May 16, I observed the larvae in the nest lying on their backs, devouring the pieces of some insect which I could not identify.

The former of these observations made in the field led me to observe the feeding of the larvae in my artificial nest of *Leptogenys*. I had frequently wondered at the way in which these ants decapitated termite nymphs or cut off their abdomens and scattered these about among their larvae. It was all quite clear to me now; examination with the lens showed that the larvae had inserted their long necks through the cut surfaces into the soft parts of the termites and were feeding exactly like the larvae of *Pachycondyla*.

During the month of May I had frequent opportunity to see *Odontomachus* feeding its larvae in my artificial nests. These larvae are placed by the ants on their broad backs, and their heads and necks are folded over onto the concave ventral surface, which serves as a table or trough on which the food is placed by the workers. The following observations are transcribed from my notebook:

semiliquide; e tale é pure in massima l'alimento delle stesse formiche allo stato adulto, quando si cibano di sostanze zuccherine vegetali o degli escrementi liquidi degli afidi. Però, molte formiche vivono pure in parte di preda, e nulla prova che si contentino di sorbire i succhi della loro vittima, e non digeriscano pure, mediante la saliva, alcune parti solide." . . . "Ora sarebbe pure possibile che Formiche, le quale vivono principalmente di preda, diano in pasto alle loro larve pezzi più o meno tritutati del corpo delle loro vittime come fanno le Vespe."

May 13. This evening several house-flies, placed in the Janet nest of *O. haematodes*, were at once shorn of their legs, then decapitated, and finally their thoraces and abdomens were cut into smaller pieces and distributed among the larvae. One was given a fly's head, which it kept twirling around in a comical manner, while it devoured the brain through the small cervical orifice. Another was given a piece of a thorax with one of the wings still attached, another a piece of an abdomen, still another, a leg with a mass of muscle at its coxal end, etc.

May 16. This evening a small homopterous insect was placed in the *Odontomachus* nest. One of the ants (A) snapped at it, disabled it, and then left it. A few moments later it was picked up by another ant (B) and carried into the chamber containing the larvae and pupae. Thereupon a third ant (C) took hold of it and began tugging at it with B till it was torn open, but not into pieces. B then placed it on the flat ventral surface of a medium-sized larva, which began feeding at once, moving the homopteron around with its jaws. After four minutes had elapsed, another ant (D) that had been standing near by, apparently much interested in the feeding, suddenly tore the morsel away and placed it on a small larva. This larva was permitted to feed ten minutes, closely watched during all this time by ant D and another (E) which had come up in the mean time. Then ant D tried to tear the morsel away from the small larva, but apparently unable to do so, it took up the larva with the morsel and dumped them both on the ventral surface of a large larva. This creature seized the homopteron and forced the small larva to release its hold and to drop to the ground. The large larva fed for fully twenty minutes, closely watched by ant D and two others (E and F). All of these ants tried at different times to wrench the morsel away from the larva, but failed. Suddenly a small ant (G) rushed up, tore it away, and ran off with it. By this time very little was left of the homopteron and I lost track of it.

May 23. A few crumbs of cake, moistened with water, were placed in the *Odontomachus* nest at 11.7 P.M. A worker soon carried one of the crumbs into the breeding chamber and gave

it to a large larva at 11.20. This larva fed but a few moments, but the cake was not removed till 11.35, when it was carried into another chamber, then at once brought back and placed between three larvae, from one of which it had just been taken. The smallest of these three larvae nibbled at it for a short time, beginning at 11.40. But one minute later this larva was carried away by a worker, and the cake was taken by another worker and given to a small larva at 11.43. This larva, too, was soon carried away (at 11.48), and the cake was taken to a large larva, which would have none of it. It was not removed, however, till 11.50. Then it was given by another worker to a large larva, which did eat some of it. At 11.51 the piece of cake, but little diminished in size after all its perambulations, was taken to another large larva. The ant remained over the larva holding the cake in place till 11.58, when another worker came up and ran away with the larva. While the larvae were feeding, the ants themselves could be plainly seen to partake of the cake from time to time. During the whole period of the above observations, and for some minutes later, *i.e.*, for over an hour, one little larva was permitted to feed without interruption on what seemed to be a piece of a house-fly.

These observations lead us to several interesting reflections. First, it is certain that the feeding of the larva of the Ponerinae is of a far more primitive character than in any other ants in which this process has been studied. It is, in fact, even more primitive than the corresponding habit of the social wasps, which feed their larvae with masticated insect prey, for in the Ponerinae the prey is cut into a few pieces only, for the purpose of exposing the soft tissues and making them accessible to the mandibles of the larvae. Myriopods or large insects are disarticulated for this purpose, small insects are merely torn open. Leaving the question of systematic affinities out of consideration, the Ponerinae may be said to have habits of feeding the young intermediate between the habits of the solitary wasps, which provide their young with whole insects, and the social wasps, which masticate the food for their larvae. In this statement it may, perhaps, be more accurate to substitute the Bembecidae for the solitary wasps, since the Bembecidae,

which feed their larvae from day to day with entire Diptera in a fresh condition, resemble the Ponerinae more closely than do the solitary wasps, which merely enclose their eggs with paralyzed larvae, spiders, grasshoppers, etc.¹ From the condition of the Ponerinae to that of the more specialized ants, which feed their larvae with nothing but the liquid food regurgitated from their own crops or from their salivary glands, the transition is very abrupt. But there are many ants whose habits have not been studied, and some of these may yet be found to bridge this chasm.

In the second place, the above-recorded observations seem to show that the Ponerine method of feeding the larvae is of a most capricious and irregular character. The quantity and quality of the food given to a particular larva, and the time it is permitted to feed, seem to be matters requiring no very strict regulation. The ants that feed the young rarely act in concert, but rather with a whimsical individualism that seems at times to border on the ridiculous.

This irregular method of feeding suggests other considerations of a wider bearing. It is generally admitted that the polymorphism of the female sex in ants, *i.e.*, the occurrence of fertile females and of sterile females of one or more casts, is in some manner correlated with the feeding of the larvae developed from fertilized eggs. In other words, the worker ants can control the production of individuals like themselves and of individuals like their queen. It is further maintained that these differences are effected by the quantity and quality of the food administered to the larvae at a certain period of their development; but here our knowledge ends. These data have been accumulated from the study of the specialized Myrmicine and Formicine ants of Europe and North America, and are

¹ Fine descriptions of wasps (Polistes) and Bembecids feeding their young are to be found in the charming works of Fabre (*Souvenirs Entomologiques*, 1^o ser., 2^m edit., Paris, 1894, pp. 126-128 and pp. 226 *et seq.*) and of Dr. and Mrs. G. W. Peckham ("The Instincts and Habits of the Solitary Wasps," *Bull. Wisconsin Geol. N. H. Survey*, No. 2, 1899, 245 pages, 14 plates). Janet has described the corresponding habits of *Vespa* ("Études sur les fourmis, les guêpes, et les abeilles." 10. Note. Sur *Vespa media*, *V. silvestris*, et *V. saxonica*, *Mém. de la Soc. Acad. de l'Oise*, tome xvi, 1895, p. 39).

supported by many valuable observations on the hive-bee. Now, while we can, perhaps, understand how these more specialized ants may manage to control the quantity and quality of liquid food regurgitated from their own crops and salivary glands, it is not so easy to understand how ants can exercise such control when they adopt a capricious method of feeding like that of the Ponerinae. Such a method can hardly produce clear-cut results; *i.e.*, either workers or fertile females. And a comparative study of the better known species of Ponerinae shows that in certain species at least there is no such sharp distinction between the sterile and fertile female as we find in the more specialized ants. Not only is the female sex in a state of morphological and physiological instability, — *i.e.*, di- or even tri-morphic, — but the male sex also is sometimes dimorphic — at least in the same genus, if not in the same species. For the purpose of illustrating this singular instability of the sexes I have compiled the following table from the literature to which I have access.¹ It includes twelve of the better known species

Species of Ponerinae.	Winged Male.	Ergatoid Male.	Winged Female.	Ergatoid Female.	Worker Major.	Worker Minor.
<i>Odontomachus haematodes</i> Linn	+		+	+	+	
<i>Pachycondyla harpax</i> Fabr. . .	+		+	+	+	
<i>Cardiocondyla Emeryi</i> Forel . .	+		+	+	+	
<i>Cardiocondyla Wroughtonii</i> Forel		+	+		+	
<i>Cardiocondyla Stambuloffi</i> Forel		+	+		+	
<i>Leptogenys elongata</i> Buck. . .	+			+	+	
<i>Ponera ergatandria</i> Forel . . .	? +	+	+		+	
<i>Ponera ochracea</i> Mayr.	+		+		+	
<i>Ponera Eduardi</i> Forel	+		+	+	+	
<i>Ponera coarctata</i> Latr.	+		+	+	+	
<i>Ponera punctatissima</i> Rog. . .		+	+		+	
<i>Stigmatomma pallipes</i> Hald. . .	+		+		+	

¹ Sharp, "Formicidae in Cambridge Natural History," *Insects*, vol. vi; Emery, "Sopra Alcune Formiche della Fauna Mediterranea," *R. Accad. delle Scienze dell' Istituto di Bologna*, 21 Apr., 1895; Emery, "Beiträge zur Kenntniss der nordamerikanischen Ameisenfauna," *Schluss, Zool. Jahrb., Abth. f. Syst., Bd. viii.*

of Ponerinae. The presence of a particular sexual phase is indicated by a cross.

Although it is by no means certain that the irregular polymorphism of the two sexes of the Ponerinae, as indicated in this table, is due to an inability on the part of the ants to regulate with precision the quality and quantity of the food administered to the larvae, I nevertheless believe that there is some causal connection between these two peculiar phenomena. At any rate, we may assume this connection as a working hypothesis for future experimentation and observation. I believe that continued study of the relatively undifferentiated sexual conditions of the Ponerinae may lead us more rapidly to a solution of the interesting problems of nutritional polymorphism than a study of the more specialized ants.

When the larvae of the Ponerinae are mature they are, like the mature larvae of the Formicinae, buried in the soil till they have spun their cocoons. They are then unearthed and the small adherent particles of soil are carefully removed by the workers. I have watched the burying of the larvae in *Leptogenys* and the unearthing and cleansing of the cocoon in *Odontomachus*. The cocoons of the three species of Ponerinae are usually kept together, but the ants are scarcely as careful in this respect as the species of *Formica* and *Camponotus* which I have observed (*F. neorufibarbis* and *C. castaneus*). Nor do they keep their larvae assorted according to sizes, a peculiarity which accentuates the irregularity of their feeding habits.

Forel, as we have seen, believes that *Ponera coarctata* may escape from its cocoon without the assistance of the workers. Unfortunately I had to leave my work at Austin before the pupae of *Odontomachus* were ready to hatch, but I am convinced that *Leptogenys*, at any rate, opens the cocoon and draws out the pupa when ready to enter on its imaginal life. I have not seen this operation under normal circumstances, as the two workers which appeared as callows in my artificial nest left their cocoons when I was not present, but for some reason the workers in this nest were continually opening the cocoons near one end and pulling out the still white pupae. Ten or a dozen workers would gather about

one of these extracted pupae and lick it for hours. Sometimes one ant would take possession of the limp thing and hold it astraddle for a long time. Ultimately these prematurely born ants were either devoured by the workers or fed to the ravenous larvae. Nevertheless the deft manner with which the cocoon was opened, the pupa extracted, and the empty cocoon at once placed on the kitchen-midden, or rubbish heap, indicated very clearly that this is also the method of procedure with pupae that have reached their full growth.

A word in conclusion concerning myrmecophiles, for the Ponerinae, like the other subfamilies of ants, are known to harbor arthropod guests in their nests.¹ No guests were taken with *Odontomachus* and *Leptogenys*, but some six different species were observed in various nests of *Pachycondyla harpax*. Only one of these had not previously been found in the nests of other species of ants near Austin. This was a small yellow ant, a *Solenopsis*, allied to the European *S. fugax*, and found inhabiting some very minute galleries in the earth between the huge burrows of the Ponerine. It is probably a "Diebsameise," given to myrmecoclepsy like its European

¹ The Ponerine guests enumerated by Wasmann in his *Kritisches Verzeichniss der myrmekophilen und termitophilen Arthropoden*, Berlin, 1894, are the following: *Typhlopemys hypogaea* Rey (staphylinid beetle), with *Typhlopone oraniensis* Luc., Palestine; *Apocellus* (?) *sphaericollis* Say (staphylinid), with *Ponera coarctata* Latr., North America; *Mesotrochus paradoxus* Wasm. (staphylinid), with *Typhlomyrmex Rogenhoferi* Mayr, Santa Catharina; *Euplectus Sikorae* Wasm. (pselaphid beetle), with *Ponera Johanna* Forel, Madagascar; *Trichonyx sulcicollis* Rchbch. (pselaphid), with *Ponera coarctata* Latr., Europe; *Amauronyx Märkeli* Aubé (pselaphid), with *Ponera coarctata* Latr., Switzerland; *Araniops amblyoponica* Brend. (pselaphid), with *Stigmatomma pallipes* Hald., Pennsylvania, North Carolina; *Tmesiphorus formicinus* McL. (pselaphid), with *Ectatomma sociale* McL., Australia; *Leptotrichus inquilinus* Koelbel (isopod crustacean), with *Ponera senarensis* Mayr, East Africa. More recently Wasmann has described the following ponerinaphiles: *Fauvelia permira* Wasm. (staphylinid), with *Pachycondyla Fauveli* Emery, Bolivia ("Die Ameisen- und Termitengäste von Brasilien," 1. Theil, *Verhandl. d. k. k. zool. bot. Gesell.*, Wien, Jahrg. 1895, pp. 40, 41); *Lomechon Alfaro* Wasm. (silphid), with *Pachycondyla aenescens* Mayr, Costa Rica ("Ein neues myrmekophiles Silphidengenys aus Costa Rica," *Deutsch. Ent. Zeitschr.*, Heft. ii, 1897); *Myrmedonia lobopeltina* Wasm. (staphylinid) and *Demera Fauveli* Wasm. (staphylinid), with *Leptogenys (Lobopelta) nitida* Sm., Natal ("Zwei neue Lobopelta-Gäste aus Südafrika," *Deutsch. Ent. Zeitschr.*, Heft. ii, 1899).

congener. Two specimens of the very singular little ant, *Strumigenys louisianae* Rog., were also taken from the earth of this same nest. Their relations with the *Pachycondyla* were probably of a more accidental nature. The other forms taken are pleomyrmecophilous, *i.e.*, they occur in the nests of several other species of ants in the vicinity of Austin. These are, first, a yellowish white species of *Lepismima*, quite common in the nests of *Pachycondyla*, but even more abundant in the nests of *Camponotus castaneus* Latr., in the same localities. This Thysanuran was also taken in the nests of *Eciton coecum* Latr. Second, a white Collembolan, similar to, if not the same as, *Cyphodeira (Beckia) albinos* Nicol. of Europe. This insect is panmyrmecophilous, occurring in the nests of nearly all the ants of Travis County. Third, *Myrmecophila nebrascensis* Bruner, rare in the nests of *Pachycondyla*, but very common in the nests of *Formica fusca*, var. *neorufibarbis* Mayr. I have no doubt that this singular little cricket had strayed from the *Formica* to the *Ponerine* nests. Fourth, a small Trichopterygid beetle was sometimes found in the nests of *Pachycondyla*. As this same species was very common in the nests of *Camponotus castaneus*, in the same localities, I believe that it, too, may have strayed from the nests of its typical host.

UNIVERSITY OF TEXAS MEDICAL SCHOOL,
GALVESTON, June 10, 1900.

THE EYES OF THE BLIND VERTEBRATES OF NORTH AMERICA. III.

*THE STRUCTURE AND ONTOGENIC DEGENERATION OF THE
EYES OF THE MISSOURI CAVE SALAMANDER, AN
ACCOUNT BASED ON MATERIAL COLLECTED
WITH A GRANT FROM THE ELIZABETH
THOMPSON SCIENCE FUND.¹*

CARL H. EIGENMANN AND WINFIELD AUGUSTUS DENNY.

A SINGLE specimen of a salamander was discovered in Rock House Cave, Barrie County, Missouri, by Mr. F. A. Sampson in July, 1891. The specimen was described by Stejneger in the *Proceedings of the U. S. National Museum*, Vol. XV, p. 115, as *Typhlotriton spelæus*. His diagnosis reads as follows: "Vertebrae opisthocoelous; parasphenoid teeth; vomerine teeth; eyes concealed under the continuous skin of the head; tongue attached in front and along the median line, free laterally and posteriorly; maxillar and mandibular teeth small and numerous; vomerine teeth in two strongly curved series; parasphenoid patches separate; nostrils very small; toes five; sixteen costal grooves, or eighteen if counting the axillary and groin grooves; tail slightly compressed, not finned; toes nearly half webbed; vomerine teeth in two V-shaped series with the curvatures directed forward; gular fold strong, very concave anteriorly; color uniformly pale."

Stejneger fully appreciated the value and nature of his discovery. He says: "Although many of our salamanders are known to inhabit caves, this seems to be the only one, so far discovered, which, like some of the other animals exclusively living in caves, has become blind or nearly so." This was written by him before he discovered the *Typhlomolge* in the underground streams of Texas.

¹ *Contribution from the Zoölogical Laboratory of the Indiana University, No. 31.*

A preliminary note by the present authors (*Proc. Ind. Acad. Sci.*, 1898, p. 252, 1899) completes the list of papers dealing with this species.

In the spring of 1897 Dr. Eigenmann visited Rock House Cave and secured a number of larvae, which Dr. Stejneger pronounced the larvae of *Typhlotriton*. Later Mr. E. A. Schultze informed him that he had seen this salamander in the underground passage to Blondi's Throne Room in Marble Cave, Stone County, Missouri. In September of 1898 he visited this cave and secured four adults and three larvae of *Typhlotriton*. A large number of the larvae were obtained from Rock House Cave a few days later. Those from the latter cave were found under loose stones and gravel in the rivulet at the mouth of the cave. They had been exposed to the light. It is scarcely supposable that those from Marble Cave had ever been affected by the light. In the caves both larvae and adults are found under the stones, the old ones in and out of the water. Occasionally one is seen lying on the bottom of a pool.

In the aquarium the larvae creep into or under anything available; a glass tube serves as a "hiding" place. The rubber tube admitting water to the aquarium is sometimes occupied by several during a temporary cessation of the flow of water. A wire screen sloping from the bottom of the aquarium formed the most popular collecting place for the larvae. They collected beneath this, although it was no protection from the light. From these observations it seems probable that stereotropism rather than negative heliotropism accounts for the presence of this species in the caves, and that this reaction has been retained after the long stay of the species in caves necessary to account for the changes in its eyes.

The eyes of the larvae when examined from the surface appear perfectly normal, but they are little used in distinguishing objects. When hungry they will strike at a stick held in the hand as they would at food. A stick lying at the bottom of the aquarium undisturbed is not molested. They strike at a worm when touched by it, or when it approaches close enough for its motion to be perceived.

In the larvae up to 90 mm. long the skin passes over the

eye without forming a free orbital rim and the eye does not protrude beyond the general contour of the head. In the adult from 97 mm. on, the eye forms a bead-like projection. There are in the adult distinct lids. These are closed over the eye, covering it entirely, the slit being much too small for the eye. The lower lid is free from pigment, but the upper lid, which closes over the lower, is as thickly pigmented as any other part of the body.

Stejneger says of the eyes that they are "small, only slightly raised, and covered by the continuous skin of the head, with only a shallow groove to indicate the opening between the lids, the underlying eyes visible as two ill-defined dusky spots."

In sections the lids are seen to overlap one another some distance, forming an obscure, free orbital rim. Fig. 1, *a*, is a median section of the lids and corneal epithelium of an eye .954 mm. in diameter, taken from an adult specimen 106 mm. in length. In this section the upper lid overlaps the lower lid .216 mm., or more than one-fifth the diameter of the eye. Passing from the median section toward the corners of the eye, the lower lid unites with the underlying tissue first. When observed from the top the upper lid covers the entire eye. The orbital slit is .17 mm. in length. The conjunctival pocket extends some distance forward and backward beyond the slit. The eye increases in size but little from the larval to the adult stage, and its growth is not proportional to the growth in length of the animal. (See comparative measurements of the eyes at the close of the paper.)

The following is a series of measurements on the larvae of *Typhlotriton*.

	ROCK HOUSE CAVE.	ROCK HOUSE CAVE.	MARBLE CAVE.
Specimen . . .	54 mm. long.	78 mm. long.	88 mm. long.
Size of pupil . .	.432 mm.	.640 mm.	—
Length of eye . .	1.30 mm.	1.50 mm.	1.60 mm.
From optic nerve			
to front of lens .	.80 mm.	1.20 mm.	—
Vertical diameter	—	1.248 mm.	1.28 mm.

Sections of the adult and larva from Marble Cave were made in the usual manner. The six normal eye muscles were pres-

ent in *Typhlotriton*. The m. recti form a sheath about the optic nerve in its distal part and spread out from it near the eye. In the adult the sclera is a layer of uniform thickness except in the region of the entrance of the optic nerve. It is not usually separated from the adjoining parts of the eye, but in places is retracted a short distance from the choroid coat by the action of reagents. It is for the most part fibrous, with few compressed nuclei, and varies from 18 to 40μ in thickness. In the larva a narrow cartilaginous band surrounds all but the ventral wall of the eye. In a specimen 35 mm. long the width of the band is about 30μ , its thickness 16μ . In three adult specimens the sclera of only one had any traces of cartilage. In the right eye of the adult specimen 103 mm. long a cartilage about 36μ thick, 60μ wide, and not more than 40μ long is found on the upper face of the eye. The absence of this cartilage in the adult has probably no connection with the degeneration of the eye. Its presence is probably a larval characteristic which disappears as the gills disappear during the metamorphosis.

The average thickness of the cornea is 40μ . In the adult it is covered by a layer of stratified epithelium, 25μ in thickness, consisting of three rows of cells. The cells of the inner row are columnar in shape, those of the middle row rounded, and those of the outer row are very much flattened and elongated (Fig. 1, *a*).

In the adult the choroid coat is usually separated from the pigment layer, but adheres closely to the sclera. In general it is thicker at the back part of the eye, and quite decidedly so at the entrance of the optic nerve. The lens is normal. Its size is given in the table at the end of the paper.

The layers of the retina are well developed in the larva. The retina of the larva differs from that of an *Amblystoma* larva in the greater thickness of its ganglionic layer. This layer is, in the young larva of *Typhlotriton*, composed of five or six layers of cells. This thickness may in part be an artifact, since the retinae examined are shrunk away from the pigment epithelium, and the ganglionic layer is in contact with the lens. In the larva 90 mm. long this layer has been reduced

to not more than three series of cells. Aside from the differences noted above, the eye of the larvae of *Typhlotriton* is apparently normal in all of its histological details. This relative thickness in the different sizes of the larvae may be gathered from Figs. 2-5 and from the comparative table at the end of the paper.

Figs. 2-5 are drawn with the same magnification and show the relative thickness of the different layers in the retinae of the larvae of different sizes and of the adult. The adult retina is reduced in thickness by the absence of the rods and cones and the (partial?) atrophy of the outer reticular layer and by the thinning of the ganglionic layer. The ganglionic layer in the adult contains from two to five rows of cells. In this respect, the adult approaches the condition found in *Amblystoma* more than the young does. The inner reticular layer is comparatively thick, that of the young being thicker than that of the adult.

In the adult the inner nuclear layer is continuous with the outer nuclear layer. (See Fig. 5.)

The inner nuclear layer consists of about seven series of cells in the smallest larva and of four to seven in the largest. The cells in the preparations available cannot be separated into bipolar and spongioblastic layers, nor are horizontal cell layers distinguishable. The outer reticular layer is well differentiated but quite thin in the larvae, and is irregular in outline, adapting itself to the overlying nuclei which encroach on its outlines. In the adult this layer is indistinguishable by the same methods that make it conspicuous in the larva. In places there appeared an open space where the outer reticular layer should be (Fig. 9), but none of its structure remains. It is fair to suppose that the fibers forming this layer are resorbed during the metamorphosis. This layer seems to be the very first obliterated by the processes of degeneration both ontogenic and phylogenetic in this as in other vertebrates with a degenerating eye.

The greatest change during and shortly after metamorphosis takes place in the layer of the rods and cones. In the larva 35 mm. long, from the mouth of Rock House Cave, the rods reach an extreme length of 50μ . The relative sizes and

number of these as compared with the much smaller cones may be gathered from Fig. 2, *a*.

In the larva 90 mm. long the outer segments of the rods are much shorter and stain less conspicuously than in the younger. The nuclei of the outer nuclear layer are distinctly in two layers, whereas in the younger they are in three less regular layers. The cones are correspondingly fainter than in the young. It is surprising that whereas in the larva 90 mm. long we find the rods and cones well developed they have greatly degenerated or practically disappeared in the adult only a few mm. longer. In an adult specimen 97 mm. long the rods have retained their normal shape and position, but I have not been able to detect any differentiation into inner and outer segments. In longer ones most of the nuclei of the outer series have become rounded at both ends. But one cone was found in eyes of the adult over 100 mm. long. It is shown in Fig. 6. In an adult specimen 103 mm. long filmy rods are still evident. They appear as conical spaces above the nuclei free from pigment rather than as possessing any demonstrable structure. Just at the margin of the place where the pigment has been torn from the retina one of these is drawn out to a great length. The pigment in this individual extends in places down between the cells of the cones. This latter condition appears in a very exaggerated form in the eye of *Typhlomolge*. In tangential section this condition and the filmy rods give rise to the appearance represented in Fig. 5, *a*.

Distinct signs of ontogenic degeneration are also seen in other parts of the retina. For instance, many nuclei of the inner series of the outer nuclear layer are shriveled. In some eyes the ganglionic nuclei have for the greater part lost their granular structure and show a homogeneous pasty condition, only a few cells with granular nuclei being present (Fig. 5). The same is true in large part of the inner nuclei of the inner nuclear layer. This condition of the ganglionic nuclei is not entirely confined to the adult but is also found in the larva.

Some of the modifications in the shapes of the outer nuclei in the adult are shown in the figures. In Fig. 7 the upper

portion of the nucleus is very much elongated. This form is of frequent occurrence. In Fig. 8 is shown the common form where the nuclei are simple elliptical bodies, which give no evidence whatever of any processes uniting them with the other elements of the retina. The Müllerian fibers are profusely present and of very large size in both larva and adult. In both adult and young the optic nerve enters as a single strand and passes entirely through the layers. A heavy mass of pigment is found following the optic nerve to within a short distance of the brain.

AVERAGE MEASUREMENTS OF THE EYES OF *TYPHLOTriton*.

LENGTH OF SPECIMEN.	35 mm.	48 mm.	62 mm.	90 mm.	97 mm.	103 mm.	106 mm.
Vertical diameter of eye	810	800	—	960	—	800	1170
From front of lens to back of eye . .	600	672	—	720	720	720	1134
Outer nuclear layer with the rods . .	76	42	112	36	28	28	—
Outer reticular layer	1	2	—	—	—	—	—
Inner nuclear layer	76	72	80	50	48	72	72
Inner reticular layer	16	20	16	24	8	8	13
Ganglionic layer	68	56	64	32	24	26	26
Pigment layer	4	16	—	—	8	20	22
Optic nerve	20	25	—	—	—	23	29
Lens	342	300	—	500	432	430	504

SUMMARY.

Typhlotriton is an incipient blind salamander living in the caves of southwestern Missouri. It detects its food by the sense of touch without the use of its eyes. It is stereotropic. Its eyes show the early stages in the steps of degeneration from those of salamanders living in the open to those of the degenerate *Typhlomolge* from the caves of Texas. The lids are in process of obliteration, the upper overlapping the lower so that the eye is always covered in the adult. The sclera possesses a cartilaginous band in the larval stages but not in the adult. The disappearance of the cartilage is probably an incident of the metamorphosis, not of the degeneration the eye is undergoing. The lens is normal. The retina is normal in

the larva with a proportionally thicker ganglionic layer than in the related epigæan forms. Marked ontogenic degenerations take place during and shortly after the metamorphosis. *a.* The outer reticular layer disappears. *b.* The rods and cones lose their complexity of structure, such as differentiation into inner and outer segments, and finally are lost altogether.

EXPLANATION OF FIGURES.

All drawings were made with the aid of the Abbe camera from sectioned balsam preparations. The comparative measurements (p. 39) furnish the key to the magnification:

- | | |
|-------------------------------|-------------------------------|
| <i>ps.</i> palpebra superior. | <i>pi.</i> palpebra inferior. |
| 1. pigment epithelium. | 2. rods and cones. |
| 3. outer nuclear layer. | 4. outer reticular layer. |
| 5. horizontal cell layer. | 6. inner nuclear layer. |
| 7. spongioblastic layer. | 8. inner reticular layer. |
| 9. ganglionic layer. | 10. optic fibers. |

FIG. 1. Diagrammatic representation of the eye drawn to scale.

FIG. 1, *a.* Vertical section through the cornea and lids of an adult.

FIG. 2. Section of the retina, exclusive of pigment cells, of a larva 35 mm. long.

FIG. 2, *a.* Tangential section through the rods and cones about on a level with the innermost extent of the pigment which is seen on the right, showing the relative sizes and abundance of the rods and cones.

FIG. 3. Section of the retina of a larva 48 mm. long.

FIG. 4. Section of the retina of a larva 90 mm. long.

FIG. 4, *a.* Tangential section showing the rods and cones at about the inner limit of the pigment which is seen on the left.

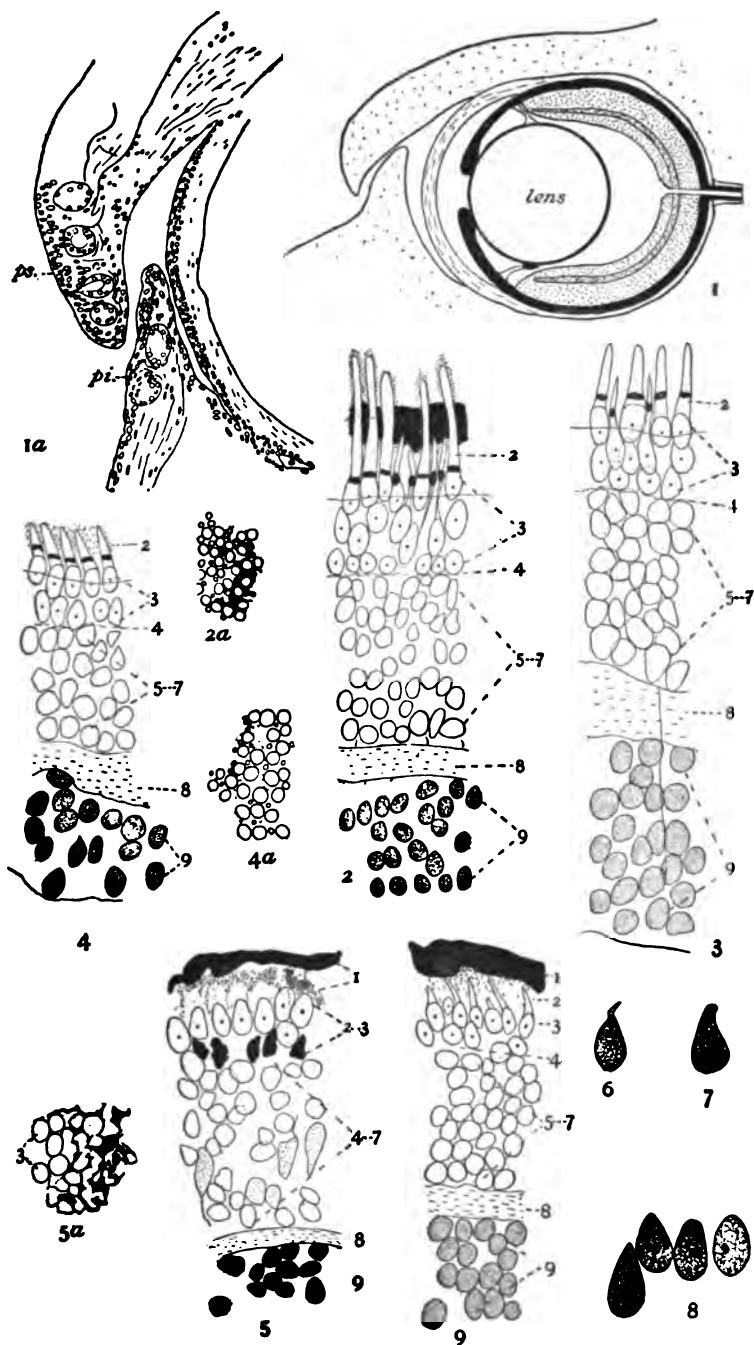
FIG. 5. Section of the retina of an adult 106 mm. long.

FIG. 5, *a.* Tangential section at about the inner limit of the pigment.

FIG. 6. The only cone found in the eyes of adults.

FIG. 7, 8. Difference in the shape of the outermost series of cells in the outer nuclear layer.

FIG. 9. Section of the retina of an adult 97 mm. long.



BIOLOGICAL BULLETIN.

THE HABITS OF PONERA AND STIGMATOMMA.¹

WILLIAM MORTON WHEELER.

IN a recent number of the *Biological Bulletin*² I described the habits of three Texan ants belonging to the subfamily Ponerinae. During the past summer an excellent opportunity presented itself to extend these observations to two other forms widely distributed in the Eastern and Northern States, viz., *Ponera coarctata* Latr. and *Stigmatomma pallipes* Hald. These are of no little interest to the student of ant life, the former as a member of the typical genus, the latter as the only known North American representative of the most primitive tribe (Amblyoponi) of the subfamily. European myrmecologists have long wished to gain some knowledge of the habits of *P. coarctata*, but its rare and local occurrence on their continent has rendered this impossible up to the present time. The European type of the genus *Stigmatomma*, *S. denticulatum* Roger, is also rarely seen, and for the same reason its habits are all but unknown.

As both the ants to be considered in this paper are subterranean and very timid, it is impossible to learn much about them in their natural environment. It is therefore necessary to keep them in artificial nests. This is, fortunately, an easy matter, since the ants are very hardy. As the colonies are small, it suffices to use for this purpose the Petri dishes employed by

¹ *Contributions from the Zoölogical Laboratory of the University of Texas*, No. 10.

² "A Study of some Texan Ponerinae," *Biol. Bull.* Vol. ii, No. 1, pp. 1-31. October, 1900.

bacteriologists for growing cultures of micro-organisms. The ants are hastily scooped up, together with their larvae, pupae, and much of the earth in which they have excavated their nest, and the whole is transferred to a Petri dish. One or two glass slides are then placed on the earth, which is spread out till it forms a layer not more than about 5 mm. in thickness. The Petri dish is kept covered to retain the moisture in the soil. In the course of a day or two the ants excavate rough-walled chambers under the slide and galleries in the adjacent soil, of the same size and shape as those which they are in the habit of forming in their natural nests. They also gather their eggs, larvae, and pupae into these chambers, where they may be easily seen. When the slides become smeared or covered with earth they can at any time be hastily replaced by clean ones without greatly disturbing the ants.

The Ponerinae may appear to lead very monotonous lives to any one who has kept under observation the different species of *Myrmica*, *Pogonomyrmex*, *Lasius*, *Camponotus*, and *Formica*. But this very monotony is full of interest to the observer who sees in the rudimental activities of these ants a certain picture, however imperfect, of the simple stages through which the higher ants have passed in attaining to their present remarkably differentiated social organizations. It can hardly be doubted that there is a phylogeny of instincts, as there is a phylogeny of structures, and there is certainly no single animal group which more clearly illustrates the truth of this statement than the Formicidae.

PONERA COARCTATA LATREILLE.

Our American *P. coarctata* is considered by Emery¹ to differ sufficiently from the European form to be ranked as a subspecies, which he calls *pennsylvanica* Buckley. In the worker the single node forming the pedicel of the abdomen is somewhat thicker and much broader behind and less narrowed anteriorly than in the European forms. The punctation of

¹ "Beiträge zur Kenntniss der nordamerikanischen Ameisenfauna" (Schluss), *Zool. Jahrb.* Abth. f. System. Bd. viii, pp. 257-360, Taf. VIII. 1894.

the head is finer, that of the thorax and node much denser and more distinct. Emery also mentions some differences in the neururation of the wings of the male: "in den Flügeln verbindet sich aber die *Costa recurrens* etwas weiter von der Gabelung mit dem hintern Ast der *Costa cubitalis*, ungefähr wie bei der europäischen *P. punctatissima*."

Figs. 1, 2, and 3, from camera drawings, represent the outlines of the male, female, and worker of the American *coarctata*. The eyes of the worker are minute and vestigial, those of the female considerably and those of the male very much larger. The worker has no ocelli; those of the female are small, while in the male they are very prominent. The node in the male and female is more slender than that of the worker, and of a somewhat different shape. The antennae of the male are of nearly uniform thickness throughout and 13-jointed, whereas the geniculate antennae of the female and worker are 12-jointed, with a long basal joint, or scape, and a club-shaped funicular portion, with much shortened middle joints. The worker and female are provided with a long sting; while the pygidium of the male ends in an acute point.

The coloration of the female and worker is highly variable. Typical specimens have the head, thorax, node, and base of the abdomen black, the mandibles, clypeus, frontal carinae, antennae, legs, posterior third of the first abdominal segment, and the tip of the abdomen from the base of the fourth segment, red or yellow. Very often the ventral portions of the trunk are more or less suffused with red or yellow, especially when the specimens are immersed in alcohol. Some specimens, probably more or less immature, are red or yellow throughout. The body is covered in all cases with short pale pubescence, which on the head forms two lines, one on either side running parallel with the straight lateral edges. These lines are apparent only in dry specimens seen in a certain light. The male is black, with the palpi, trochanters, knees, tips of tibiae, and the tarsi light yellow. The genitalia and the incisures of the segments of the slender abdomen are also more or less yellow or piceous, as are also the stigma and veins of the colorless wings, both in this sex and in the female.

P. coarctata is a small ant, the male and female measuring scarcely more than 4 mm. in length, while the workers vary from 3 to 3.75 mm.

According to Emery this ant occasionally presents ergatoid females. He mentions¹ two of these wingless individuals from Sicily, with eyes somewhat larger than those of the worker, with ocelli and with the node somewhat higher and more slender above. I have been unable to find any such specimens among

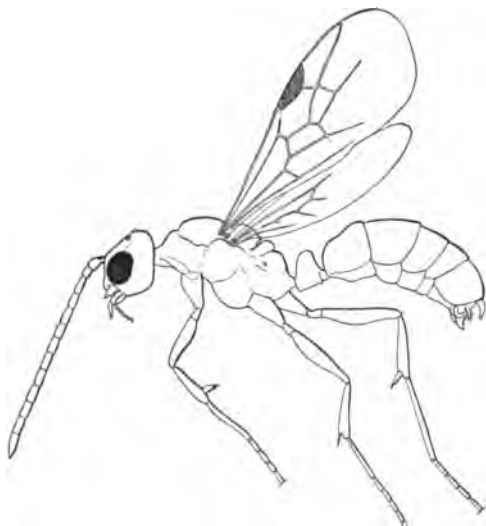


FIG. 1. — *Ponera coarctata* Latr., subsp. *pennsylvanica* Buckl. Male.

my American material, although I carefully scrutinized no less than two hundred wingless individuals from widely separated localities and from at least twenty different nests.

P. coarctata is the most widely distributed of the European Ponerinae and occurs even in northern Africa (Algiers), according to Emery.²

In this country, too, its subspecies, *penn-*

sylvanica, is one of the most widely distributed forms in the subfamily. Emery³ has examined specimens from Pennsylvania, New Jersey, Virginia, Maryland, Mississippi, Florida, and Ohio. Forel has observed it in North Carolina,⁴ and I can add to this list four other states, *viz.*, Wisconsin, Illinois, Massachusetts, and Connecticut. It may, I think, be safely said to inhabit all the states east of the Mississippi, as well as Canada.

¹ "Sopra Alcune Formiche della Fauna Mediterranea," *Mem. letta alla R. Accad. delle Scienze dell' Istituto di Bologna*. Pp. 1-19, Tav. I. 21 Aprile, 1895.

² *loc. cit.*, p. 6.

³ "Beiträge zur Kenntniss," etc., *loc. cit.*, p. 268.

⁴ *Annales de la Soc. Entomol. de Belgique*. Tome xliii, pp. 438-447. 1899.

It is undoubtedly far more common in this country than in Europe. In July I found numerous nests at Rockford, Ill., both under the bark of old logs and under stones along the streets of the town.

It is not uncommon in similar locations at Woods Holl, Mass., and very abundant under stones on the slopes of Mt. Pisgeh (altitude 1450 feet), at Colebrook, Litchfield County, Connecticut.¹

¹ This last locality, together with the slope of a small neighboring hill, is a rich collecting ground for ants. I give here the complete list of the forms taken there during August, as it probably embraces nearly all the species of Formicidae that occur in New England: *Brachymyrmex Heeri* Forel, subsp. *depilis* Emery; *Lasius niger* L.; *L. flavus* L.; *L. umbratus* Nyl., subsp. *mixtus*, var. *aphidicola* Walsh; *L. latipes* Walsh; *Formica sanguinea* Latr., subsp. *rubicunda* Em.; *F. exsectoides* Forel, var. *opaciventris* Em.; *F.*

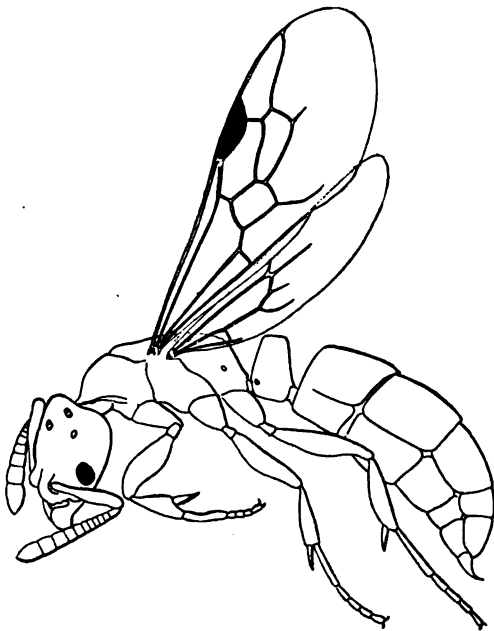


FIG. 2. — *Poner a coarctata* Latr., subsp. *pennsylvanica* Buckl. Virgin female.

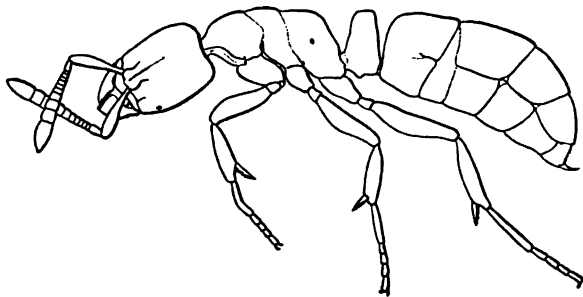


FIG. 3. — *Poner a coarctata* Buckl., subsp. *pennsylvanica* Buckl. Worker.

pallide-fulva Latr., subsp. *Schaufussi* Mayr; *F. pallide-fulva*, subsp. *nitidiventris* Em.; *F. fusca* L., var. *subsericea* Say; *F. fusca*, var. *subaenescens* Em.; *F. fusca*, subsp. *subpolita* Mayr, var. *neogagates* Em.; *Camponotus herculeanus*, subsp. *ligniperdus* Latr., var. *novaeboracensis* Fitch (= *pictus* Forel); *C. herculeanus*, subsp. *pennsylvanicus* de Geer; *Stigmatomma pallipes* Hald.; *Poner a coarctata* Latr.,

P. coarctata is not found in deep woods or in damp places, but prefers rather dry localities more or less open to the sunlight. The margins of woods and along stone walls are favorite haunts, under stones rather deeply imbedded in a rich soil, especially leaf mold. Here it excavates a small, irregular chamber, from which a few straggling burrows run off into the neighboring soil. In some cases the chamber and burrows are found under the lower surface of the stone, but I have gained the impression, from the examination of many nests, that the ants often prefer the vegetable mold nearer the surface, where it overlaps the sides of the stones. Chambers and galleries of the same irregular pattern are excavated in the rotten wood when the ants nest under the bark of old logs. The larvae and pupae are reared in the chambers. In late June and early July the nests contain eggs and larvae but no pupae; during the latter half of July and the month of August only cocoons are found, usually crowding the chamber so that the ants have little space in which to move over and among them. The imagines begin to hatch during the last two weeks of August and the first week of September. Even by the latter date I have seen no eggs nor larvae to represent a second brood.

The number of individuals composing a colony varies in different nests and with the advance of the summer. As the ants are very timid and at once seek refuge in their galleries as soon as the stone that covers the nest is moved, it is not easy to determine their precise numbers. None of the nests opened at Rockford, Ill., July 1, contained more than eight or ten ants, including a single female. As soon as the cocoons begin to hatch, the colony increases rapidly. One rather typical nest, opened at Colebrook, Conn., August 24, contained six males, one female (with wing stumps and evidently the mother of the colony), one callow virgin female, twelve

subsp. *pennsylvanica* Buckl.; *Myrmicina Latreillei* Curt., subsp. *americana* Em.; *Formicoxenus nitidulus* Nyl.; *Solenopsis molesta* Say; *Crematogaster lineolata* Say, var.; *Stenamma (Aphaenogaster) fulvum* Rog., subsp. *aquia* Buckl., var. *piceum* Em.; *Myrmica rubra* L., subsp. *scabrinodis* Nyl., var. *Schencki* Em.; *Tapinoma sessile* Say.

workers, and forty-four cocoons. A few nests examined somewhat later in the month contained a greater number of individuals, so that fifty to sixty is perhaps not too great an estimate for a large colony by the first week in September.

The winged males undoubtedly leave the nests like the males of other ants, as I have taken them in the sweep-net in the grass while collecting small Diptera. I have also seen the males copulating with the newly hatched females in the same nest. The small size of the nests in the early summer would seem to indicate that the large number of workers in the late summer and early autumn must split up into several detachments, each with a young queen, and migrate to different localities. My reasons for making this statement, apart from the above-mentioned mating of the young queens within the parental nest, are largely of a negative character, but they may be given for what they are worth. First, I have observed no tendency in the young queens, while they possess wings, to leave the nests like the males; second, the wings are often lost very soon after hatching, sometimes before the queen has acquired her deep adult coloration; and, third, I have never found a solitary queen in the act of founding a nest, either of this or of any other of the five species of Ponerinae I have studied, although I have frequently seen the young fertilized queens of *Camponotus*, *Formica*, *Lasius*, *Tapinoma*, *Crematogaster*, *Stenamma*, *Myrmica*, and *Pogonomyrmex* starting their colonies. The fact that the colonies seem to be annual instead of perennial growths, as among other ants like those above mentioned, is of considerable interest. It points to very primitive conditions in the Ponerinae, especially as the same is also true of tropical forms like *Pachycondyla* and *Leptogenys*, which can hardly be destroyed by severe winters. Thus what was at one time erroneously supposed to be true of the more specialized ants, *viz.*, the founding of a colony by a young female leaving the parental nest like the young queen of the hive bee, accompanied by a number of workers, may prove to be the normal method of nest formation with the Ponerinae. If this supposition is correct, there must be considerable inbreeding in the colonies of these ants, as the females would be regularly

fertilized by males from the same nest. There may be some connection between this condition and the limited productivity of these ants, and the strong tendency to parthenogenesis seen in some of the species (*e.g.*, in the ergatoid females of *Pachycondyla harpax*).

The behavior of *P. coarctata* towards individuals of the same species from different nests is very similar to that observed in *Pachycondyla*. If two nests be thrown together into the same dish, there may be no immediate signs of hostility; but after a few hours have elapsed, the ants are found struggling together in pairs. They interlock mandibles or tug at each other's legs and antennae, or even wrestle fiercely, intertwining their long bodies and trying to use their slender stings. These contests may be renewed from time to time for many days, whenever two individuals from different nests happen to meet, but deaths are rare, and ultimately the colonies fraternize completely. Long before the ants have settled their various difficulties, however, the cocoons and larvae of both nests are brought together as common property. A dozen different nests can be compounded quite as easily as two, and a few ants from one nest can be induced to adopt a large number of cocoons and larvae taken from half a dozen different nests.

The eyes of the workers of *P. coarctata* are so very small that they can hardly be of much service as visual organs. The actions of the ants indicate that they are guided very largely by their extremely sensitive antennae. They are, undoubtedly, able to detect the difference between light and darkness, but the fact that they do not seem to mind exposure to the light, provided they are covered with a glass plate, leads me to infer that they are rather positively stereotropic than negatively heliotropic. Of course their preference for an atmosphere charged with a certain amount of moisture—their positive hygrotopism—leads them to seek refuge in dark places, under stones or the bark of old logs.

I have not been able to ascertain the food of these ants in a state of nature. They probably kill and imbibe the juices of very small subterranean insects. In captivity they are omnivorous, feeding readily on raw or boiled meat, yolk of eggs,

corn bread, or even on "Boston brown bread." They do not appear to share the fondness of ants in general for sugar dissolved in water. When kept for a time without food they eat their dead companions or their own eggs, larvae, and pupae.

The workers of *Ponera* are never seen feeding one another with regurgitated food, like the different species of *Formica*, *Lasius*, and *Myrmica*. Even the queen is obliged to feed herself. The workers bestow on her no special attentions, nor does she enjoy any of the privileges of the queens of the above-mentioned specialized genera, after they have once established their colonies. Like any one of the workers, she takes part in digging the galleries, wanders out in search of food, assists in transporting and cleansing the eggs, larvae, and cocoons, and in feeding the larvae. Although not expressly stated in my former paper, this is also true of the ergatoid females of *Leptogenys* and *Pachycondyla*. This would seem to indicate a decidedly primitive condition, since the activities of the females of the Ponerinae never pass beyond the stage exhibited by the females of the more specialized ants only while they are raising their first batch of workers.¹

In the scrupulous care of their nests, colonies of *P. coarctata* closely resemble the more specialized ants. They bury their food or any liquid or strong-smelling substance in their environment, and all refuse — dead ants, dead pupae, empty cocoons, etc. — is deposited in one corner of the nest.

The eggs of *P. coarctata* are oblong, like those of the other Ponerinae I have described (*Pachycondyla*, *Leptogenys*), and of very large size — fully .6 mm. long, or nearly as large as the thorax of the insect that lays them. The number produced at one time is, however, relatively small. Only three were deposited by one female in my nests July 20. As the larvae found in nests in early July are of very different sizes, we must assume that the queen lays a few eggs at a time,

¹ In this connection it is interesting to note that, as Janet has shown ("Nids artificiels en plâtre. Fondation d'une colonie par une femelle isolée," *Bull. Soc. Zool.*, tome xviii, p. 168, France, 1893), the female of the more specialized ants, when separated from all her workers, may return to and repeat all the activities which she displayed while founding her first colony.

probably at intervals of a few days or a week. It is quite possible that some of the workers, acting as ergatoid females, may contribute unfertilized eggs which give rise to the males that are found in nearly every colony late in August.

The larva (Fig. 4, *a*) is clearly of the Ponerine type, though differing in a few important particulars from any of the larvae

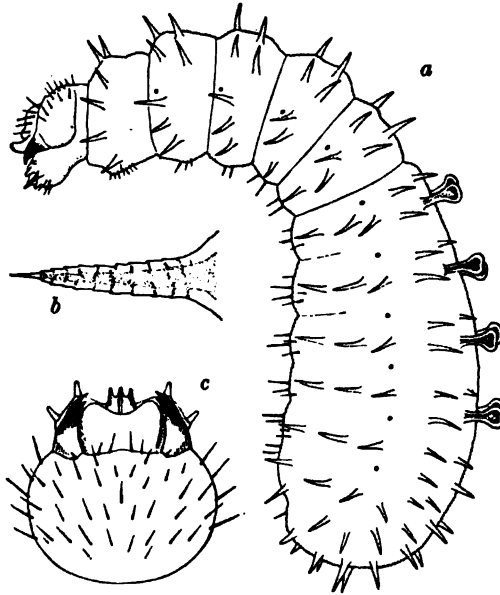


FIG. 4. — *a*, larva of *Ponera coarctata* Latr., subsp. *pennsylvanica* Buckl. Nearly ready to pupate. *b*, bristle-capped tubercle of same; *c*, head of same (dorsal aspect).

of the five genera (*Leptogenys*, *Pachycondyla*, *Ponera*, *Odontomachus*, and *Diacamma*) described by Emery¹ and myself.² It is rather robust, with a large head succeeded by five distinct segments. The remaining segments, forming the swollen abdomen, are not distinctly marked off from one another. The body is furnished with outgrowths of three different types. The

first of these is represented by a number of pointed bristles confined to the ventral surface of each segment. The second type is represented by several longitudinal rows of pointed tubercles, each of which, under a high magnification (Fig. 4, *b*) is seen to consist of a short distal spine and a long, tapering proximal base, directly continuous with the integument of the larva, and covered with transverse rows of serrated points. The distal spine is movably articulated with the proximal portion, and is so easily detached that it may be

¹ "Intorno alle Larve di Alcune Formiche," *Mem. letta alla R. Accad. delle Scienze dell' Istituto di Bologna*. Pp. 1-10, 2 Tav. 7 Maggio, 1899.

² *Loc. cit.*, pp. 15-22.

overlooked. The third type of projection is found only on the dorsal surface of the third to sixth abdominal segment as four pairs of club-shaped structures which are glutinous to the touch. That these are peculiar modifications of the tapering tubercles seems to be indicated by the fact that they replace on either side in each of the four above-mentioned segments the more posterior of the two pointed projections seen in the thoracic, first and second, and seventh and eighth abdominal segments. The larva is usually kept on its back, so that the four pairs of glutinous tubercles act as suckers and fix it to the sides of the earthen chamber or to the glass of the artificial nest. The ants have to exert a slight effort in pulling the larva away from its attachment. The head of the larva in dorsal view is shown in Fig. 4, c. It is broad, evenly rounded behind, and beset with short stiff bristles. The labrum is bilobed and does not extend beyond the tips of the powerful tridentate mandibles. The fleshy maxillae and labrum project somewhat beyond the mandibles, the former being provided with robust tactile cones, the latter with a prominent median tubercle on which opens the duct of the spinning gland. Comparison of the figures in this and my previous paper shows that the larva of *P. coarctata* is peculiar in lacking the circlets of bristles on the pointed projections and in possessing clavate adhesive tubercles on the dorsal surface of the abdomen.

The larvae are fed in the very same manner as the larvae of the large Texan Ponerinae, *i.e.*, with pieces of food and not with liquid regurgitated by the ants. In confinement I did not succeed in inducing the ants to feed their larvae with fragments of insects, but they carried crumbs of moistened corn bread to them, and the larvae could be seen lying on their backs, attached by their glutinous dorsal tubercles, slowly consuming the morsels which had been placed on their flattened ventral surfaces. The fixation of the larva to the walls of the nest seems to be an adaptation for giving freer play to the head and slender neck during feeding.

The oblong elliptical cocoons of *coarctata* are of a light buff or cream color, and vary from 2 to 3.5 mm. in length. They closely resemble the worker cocoons of *Lasius umbratus mixtus*

Nyl., var. *aphidicola*, an ant which in Massachusetts and Connecticut is often found under the same stones with the *Ponera*. The larger cocoons belong to the males and females, the smaller ones to the workers.

The eggs and larvae are looked after with great care by the ants, as Forel has observed.¹ On a former occasion, however, I expressed doubt concerning the validity of Forel's further statement: "Lorsqu'on découvre un nid de *Ponera* dans un tronc pourri, on voit leur cocons jaunes assemblés dans un coin, mais absolument abandonnés des ♀ qui n'essaient pas de les sauver, ni de les recueillir." I have since had frequent opportunity to observe these ants, and I am convinced that the master myrmecologist is in error. It is true that the slightest disturbance of the nest causes the ants to retreat into their galleries and to forsake their cocoons, but when one stops to watch the nest a few moments, one is sure to see the ants returning one by one and stealthily removing their charges. This they do rather awkwardly, walking backwards and dragging the cocoons away without lifting them from the ground, in marked contrast with *Leptogenys elongata*, which straddles the cocoon with its long legs and carries it away with surprising dexterity. Simple experiment with the artificial nests shows that the cocoons of *Ponera*, when removed to a distance of three or four inches from the chamber in which the ants have stored them, are taken back in the space of ten to thirty minutes. Nevertheless, Forel certainly deserves credit for directing attention to this matter of the care of the cocoons, for if one has observed the way in which a large and highly specialized ant, like our northern *Formica pallidefulva* Schanfussi, e.g., when its nest is uncovered, rushes out in the very face of danger to rescue its cocoons, the slow and awkward methods of *P. coarctata* certainly indicate a more primitive or possibly degenerate condition quite in harmony with the other habits of this feeble little ant. Further evidence that these ants care for their cocoons is seen in their habit of continually creeping in and out among them, and in the time which they devote to licking and cleansing them when there are no longer any larvae to require these attentions.

¹ *Loc. cit.*, p. 443.

Forel goes on to say of *P. coarctata*¹: "Je soupçonne que chez ces fourmis, moins sociales que les autres, les nymphes sortent seules de leurs cocons, sans avoir besoin de l'aide des ♂." For the purpose of testing this supposition, I tried to surprise the ants in the act of leaving their cocoons, but I was not successful, notwithstanding numerous workers, males and females, were hatching in my artificial nests. A large number of cocoons from several different nests, and apparently in a healthy condition, were isolated in small dishes, but they failed to hatch. On the other hand, the workers opened many cocoons, extracted the dead or moldy pupae, cut them up into pieces, ate portions of them, and deposited the remainder on the refuse heap. I shall show when I come to consider *Stigmatomma pallipes* that these acts, which resemble what I formerly described for *Leptogenys* and regarded as indirect proof that the living callows are assisted in their escape from the cocoons, are of no value as evidence in this matter.

On hatching, the workers of *Ponera* are light dirty yellow, and very gradually, in the course of several days, acquire their dark color. The abdomen remains pale longer than the head and thorax. The females are more mature on hatching, having the head and thorax brown. The males are quite black and fully mature soon after leaving the cocoon.

In concluding this account of *P. coarctata* a few myrmecophiles that dwell with this ant may be mentioned. In two nests, one from Rockford, Ill., the other from Colebrook, Conn., I found a small brown Pselaphid beetle. In a single nest in the latter locality a minute Staphylinid was taken. In this locality also were found some peculiar mites, often attached in pairs on either side of the node and the first abdominal segment. Their symmetrical position resembles that of the mites *Antennophorus* and *Discopoma* infesting the *Lasius umbratus mixtus* Nyl. of Europe.² Wasmann³ enumerates as

¹ *Loc. cit.*, p. 443.

² See Janet. Études sur les fourmis, les guêpes, et les abeilles. Note 13. Sur le *Lasius mixtus* l'*Antennophorus* Uhlmanni, etc. Pp. 1-62, 16 figs. Limoges, 1897.

³ Kritisches Verzeichnis der myrmekophilen und termitophilen Arthropoden. Berlin, 1894.

myrmecophiles of *P. coarctata* in North America the Staphylinid beetle *Apocellus* (?) *sphaericollis* Say, and in Europe the two Pselaphid beetles: *Trichonyx sulcicollis* Rchbch and *Amauronyx Märkeli* Aubé.

STIGMATOMMA (AMBLYOPONE) PALLIPES HALDEMANN.

Although two species of *Stigmatomma* are known from southern Europe (*S. denticulatum* Roger and *S. impressifrons* Emery), both are of rare and local occurrence. On the former species Emery¹ has published the following note: "Dr. Alessandro Fosi had the good fortune to observe the nest of *S. denticulatum* while excavating antiquities at Verucchio near Rimini. These nests were found on several occasions in the Umbrian cinerary urns, and always at the surface of the layer of ashes with bone fragments found beneath the earth which had percolated in between the lid and the original contents of the urn. It was, however, impossible to obtain the winged individuals, nor could anything further be ascertained concerning the habits of these singular subterranean ants. The population of a nest which I had occasion to examine comprised about forty individuals, three of which were females."

Our American species, too, is considered to be an uncommon insect, although it is widely distributed through eastern North America, from Canada to North Carolina.² I have found it both on the Island of Naushon, near Woods Holl, Mass., and at Colebrook, Conn. In the former locality it was very common in some rather open oak woods at the north end of the island, under large stones, imbedded in rich vegetable mold. Here, on August 6, I uncovered some thirty nests in the course of three hours. At Colebrook, after the most careful search, I succeeded in finding only three nests (August 16 to 31), and these were in widely separated localities. All the nests on Naushon Island contained great numbers of cocoons, but in only three were there eggs or larvae. One of these contained a few mature larvae ready to

¹ "Sopra Alcune Formiche," etc., *loc. cit.*, p. 3.

² Emery, "Beiträge," etc., *loc. cit.*; Forel, *loc. cit.*

pupate; the two others contained several packets of eggs and many young and half-grown larvae. One of the nests found in Colebrook, August 29, contained numerous callows and a few young larvae. These observations, together with the fact that the cocoons collected on Naushon Island nearly all hatched before August 20, show that *Stigmatomma* normally produces two broods during the summer. In this respect it may differ from *P. coarctata*; for, as I have said, no eggs or larvae of this species were found in several nests examined the last of August and the beginning of September.

S. pallipes seems to be so completely subterranean in its habits that it does not come to the surface even at night. Its nests are like those of *P. coarctata*, and it also often digs its galleries in the vegetable mold overlapping the edges instead of beneath the center of a stone. It is a much larger ant than *P. coarctata*, measuring 5.5 to 7.5 mm. One of the colonies taken on Naushon Island consisted throughout of very small individuals (5.5 to 6 mm.). The Colebrook individuals were all of smaller size than the majority of those from Massachusetts.

The females (Fig. 5), of which each colony contains from one to four before the hatching of the cocoons, are of the same size or slightly larger than the workers. They differ from the workers (Fig. 7) in having much larger lateral eyes, in having ocelli and wings, and in the structure of the thorax. Both females and workers are of a rich reddish-brown color; in older specimens the head, thorax, and node are almost or quite black, while the abdomen and legs are much paler. The male (Fig. 6) is black, with the two basal joints of the antennae, the trochanters, tibiae, and tarsi yellow; the remainder of the antennae reddish. The head, thorax, and anterior portion of the node are opaque and coarsely punctate or wrinkled, whereas the pleurae, scutellum, posterior edge of the node, and the abdomen are glabrous. The black stigma of the colorless wings in both sexes is large and conspicuous. Interesting morphological characters, such as the structure of the antennae in the two sexes, the remarkable dentate mandibles and clypeus in the female and worker, the venation, etc., are represented in

the figures and need no further description. There were no ergatoid females among the many specimens collected and reared, but it is quite possible that some of the workers may lay eggs, and, though lacking ocelli, such individuals could be called ergatoid females in

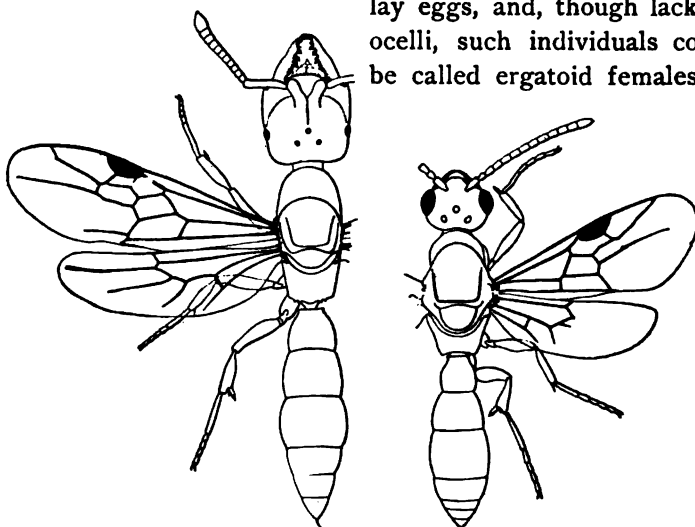


FIG. 5.

FIG. 6.

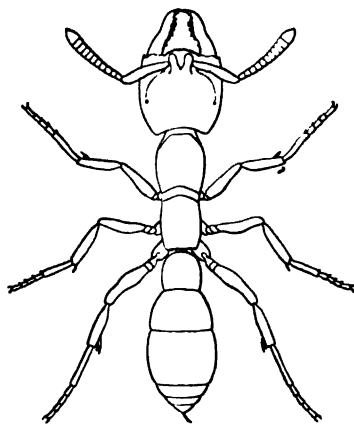


FIG. 7.

Stigmatomma pallipes Haldem. Fig. 5, Virgin female; Fig. 6, male; Fig. 7, worker.

the sense in which I have used that term (perhaps somewhat inaccurately) in my description of *Pachycondyla harpax*.¹

¹ *Loc. cit.*, p. 5.

Much of what I have said concerning the size and growth of the colonies of *Ponera* may be repeated for *Stigmatomma*. One colony taken at Colebrook contained only two workers, and another seven workers and a female; but nearly all that were collected on Naushon Island were larger, varying from ten to twenty individuals. As some of these colonies had from four to six times as many cocoons as ants, the colonies in the artificial nests by the end of August contained from forty to sixty individuals, including in some instances ten or a dozen females and as many males.

Different nests of *S. pallipes* fraternize after a struggle in much the same manner as other species of *Ponerinae*. When my supply of vials gave out, while collecting the numerous nests of this species on Naushon Island, I was obliged to put some fifteen nests into a single glass jar. There was considerable struggling among the ants of the different nests for a few days, but eventually they settled down peacefully and attended to their cocoons in common. Very few of the ants were killed in the struggle, and these were usually small individuals. A portion of this compounded colony was taken in a small dish on a day's railway journey, August 9. On arriving at my destination I found most of the ants killed, and a few that were still fighting died a few days later. The unusual severity of the struggle in this case was probably due to the close confinement of the ants in a small receptacle and the jarring to which they had been subjected for several hours. Four ants and a number of larvae taken at Colebrook, Conn., August 28, were placed in a Naushon nest which had just hatched its last cocoons. The larvae were at once appropriated by the Massachusetts ants, and later in the evening a sharp struggle ensued between the members of the two colonies. On the following morning one Connecticut ant was found dead, the three others had gone over to the enemy, and the whole colony was busy cleansing the larvae.

The eyes of the workers of *Stigmatomma* are even more rudimental than those of *Ponera*. The reactions to light and darkness, to contact and to moisture, closely resemble those of the above-described species. The females, notwithstanding

their much larger eyes and their ocelli, have the same timid, groping habits as the workers. When fully mature the males, like the males of *Ponera*, are positively heliotropic and negatively stereotropic.

Stigmatomma appears to be very sensitive to low temperatures. It passes the cold nights and mornings even during August and September curled up, with its broad head covering the tip of its abdomen. When dug out of the soil some moments elapse before it straightens and begins to run about. It probably passes the winter in the convoluted conditions.

For fully four weeks after my colonies were placed in artificial nests the ants refused to eat, although I tried a great variety of foods. When small living insects were placed in the nest they were cautiously attacked, the ant advancing, snapping at them with its long mandibles, and then retreating. This whole action was a very feeble imitation of the snapping I have described for *Odontomachus*.¹ The insects thus killed were not eaten, but covered with particles of earth. Finally the ants consented to eat some of the larvae and pupae of *Formica pallide-fulva*, and a little later they became very fond of raw meat. While feeding, the huge mandibles are kept closed with overlapping tips. The insects are obliged to slide them over the food in order to reach it with the tongue and maxillae. Like other ants, they are unable to swallow solid food, but after rasping off a small mass with the tongue, they press it in the subpharyngeal pocket, thereby extracting its juices, and then spit out the small oblong ball of residue. The whole process of forming and disposing of these "boulettes de nettoyage" is exactly like that described by Janet for *Formica rufa*, *Lasius mixtus*, and *Vespa crabro*.² The workers of *Stigmatomma* were never seen feeding one another or their queens;

¹ *Loc. cit.*, pp. 12-14.

² "Sur l'organe de nettoyage tibio-tarsien de *Myrmica rubra* L.," *Ann. Soc. Ent. de France*, tome lxiii, p. 697, 1895; "Sur *Vespa crabro*, histoire d'un nid depuis son origine," *Mém. Soc. Zool. de France*, tome viii, p. 76, 1895; Sur le *Lasius mixtus* l'*Antennophorus Uhlmanni*, etc., pp. 16, 17. Limoges, 1897.—The agricultural ant, *Pegonomyrmex barbatus* Sm., when fed on starchy substances, like rolled oats, literally sprinkles the floor and walls of its nest with snow-white "boulettes."

nor can this be readily done by ants with such huge, horizontally projecting mandibles.

Stigmatomma has a singular habit of vigorously shaking its body from time to time, as if suffering with the ague. The motion is not unlike that of the Termites when they are supposed to be stridulating. It is possible that the action of the Ponerine may produce a sound through the rubbing of the various segments and joints on one another, but, though perfectly familiar with the sounds produced by the large Myrmicines *Pogonomyrmex* and *Atta*, I have not been able to detect them in *Stigmatomma*.

What I have said concerning the habits of cleanliness in *P. coarctata* applies also to the species under consideration.

The eggs of *Stigmatomma*, though deposited by a larger insect, are smaller than those of *P. coarctata*, being only .5 mm. long. They are, how-

ever, more numerous. A packet deposited by a female in one of my artificial nests contained thirteen eggs. In shape they are oblong elliptical, like the eggs of other Ponerinae. They are not arranged with their long axes parallel with one another, as in *Pachycondyla* and *Leptogenys*, but in an irregular mass, like the eggs of all the other subfamilies of ants, including the Dorylinae (*Eciton*).

The larva has the appearance of Fig. 8, *a*. The body is rather slender in alcoholic specimens, and the segments are all quite distinct and clothed rather uniformly and densely with yellowish hairs, which under a high power (Fig. 8, *b*) are

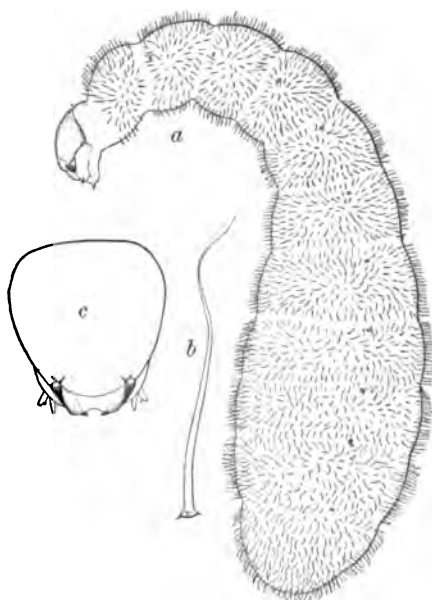


FIG. 8. — *a*, mature larva of *Stigmatomma pallipes* Haldem; *b*, bristle of same; *c*, head of same (dorsal aspect).

seen to taper into very slender flexuous points. The head is somewhat longer than broad and without hairs on its dorsal surface, the labrum is bilobed, the maxillae provided with the usual tactile cones. The outer one of these on either side appears to be bifurcate. The young differs from the mature larva only in having a relatively larger head and a sparser covering of bristles. Comparison of the larva of *Stigmatomma* with that of *Ponera*, *Pachycondyla*, *Leptogenys*, and *Odontomachus* shows that it does not conform to the Ponerine type but closely resembles, instead, the larvae of certain Myrmicinae, which are also covered with hairs instead of bristly tubercles.¹

The cocoons of *Stigmatomma* are of a slightly darker, more brownish color, and somewhat more oval than the cocoons of *Ponera*. They measure from 4.5 to 6 mm. in length.

What I have said of the care of the eggs, larvae, and pupae of *P. coarctata* is equally true of *Stigmatomma*.

The larvae are fed in the very same manner as other Ponerine larvae. In one of the nests on Naushon Island a large larva was seen with its head and neck inserted in the two last segments of a beetle larva (*Tenebrionid*), which must have been captured and disarticulated by the ants. In my artificial nests the ants carried the larvae of *Formica pallide-fulva* to their own young. The latter could be distinctly seen sucking the juices of the *Formica* larvae till they were reduced to shriveled skins. These were then carried away and placed on the refuse heap.

I have succeeded in surprising the callows in the act of escaping from their cocoons. This they do, as Forel believed to be the case with *P. coarctata*, without any assistance from the workers. Several cocoons were isolated in a watch-glass, and I had an opportunity of seeing a female, two males, and several workers emerge entirely by their own efforts. The ant gnaws through the wall of the cocoon at a spot a short distance behind the anterior pole. The shape of the incision at once indicates whether a male or a female (or worker) is

¹ Compare, e.g., my figures of the larvae of *Pogonomyrmex barbatus*, *loc. cit.*, p. 20, Fig. 9.

about to emerge. In the latter case the opening, which is produced by the huge mandibles, has the form of a transverse slit, extending halfway round the cocoon. The small, sharp mandibles of the male, however, gnaw a hole with irregular edges and of much smaller size. The insect, after periods of struggling, alternating with periods of rest, succeeds in getting first one antenna, then the other, and then the fore legs through the orifice, and finally, with considerable effort, creeps out. After making this observation on isolated cocoons I had an opportunity of making it in the artificial nests. In these the hatching cocoons were often carried about and placed on or under the stack of other cocoons, while the callows, struggling to emerge, seemed to hold out their antennae and fore legs in a supplicating attitude to the completely indifferent workers. In a few instances the callows died while halfway out of the cocoons and were carried to the refuse heap in this condition. Occasionally, when the young callows had emerged with their hind legs still enswathed and encumbered by the white pupal skin, the workers would pull this away. They also occasionally licked and fondled the newcomers, as if their bodies were covered with some pleasant secretion, but beyond these acts their helpfulness did not extend. These same workers, however, frequently opened cocoons and extracted dead immature pupae, cut them up, and then placed them on the refuse heap. This act shows that the statements concerning *Leptogenys* in my former paper¹ may require emendation.

The newly hatched *Stigmatomma*, as we should naturally expect from the above observations, is not as feeble as the callows of the more specialized ants. The males and females issue with their wings fully expanded; the former have their bodies completely pigmented and are able to run about briskly; the latter, as well as the worker callows, although of a rich yellowish-red, a color which they retain for several days, are nevertheless soon able to run about and to join in the labors of the colony. The queens show no tendency to leave the nest and usually lose their wings (after copulation?) while still in the red callow condition.

¹ *Loc. cit.*, pp. 29, 30.

Careful search in and about the natural nests of *Stigmatomma* failed to reveal the presence of any myrmecophiles. Wasmann, however, mentions¹ a Pselaphid beetle, *Araniops amblyoponica* Brend., as occurring with these ants in Pennsylvania and North Carolina.

GENERAL CONSIDERATIONS.

The foregoing observations on *Ponera* and *Stigmatomma*, together with those contained in my former paper, suggest some considerations of a more general nature.

First, the appearance of the larva of *Stigmatomma*, differing so widely from the known larvae of other Ponerinae, is calculated to raise some doubts on the subject of taxonomy. Forel² has been of the opinion that the group of genera including and allied to *Amblyopone*, viz., *Stigmatomma*, *Mystrium*, *Prionopelta*, and *Myopopone*, should be separated from the Ponerinae as an independent subfamily, the Amblyoponinae. The main characters used as an argument for this change are the presence of two spurs on the hind tibia of these ants, and the very broad and peculiar articulation of the node with the succeeding abdominal segment. Emery, on the other hand,³ has protested against raising this group to the rank of a subfamily and its separation from the Ponerinae.⁴ He argues that double spurs occur also on the hind tibiae of nearly all Ponerinae and of several Myrmicinae as well, that the articulation of the node is highly variable among the Ponerinae, and that the Cerapachyi (which Emery insists on placing with the Dorylinae) present the same conditions of the node as in *Amblyopone*. He also calls attention⁵ to the important fact that the

¹ *Loc. cit.*, p. 94.

² "Sur la classification de la famille des Formicidae," *Annales de la Soc. Entomol. de Belgique*. Tome xxxvii, pp. 161-165. 1893.

³ "Die Gattung *Dorylus* Fab. und die systematische Eintheilung der Formiciden," *Zool. Jahrb.* Abth. f. System. Bd. viii, pp. 685-778, Taf. XIV-XVII, 41 text-figs. 1895.

⁴ "Die Abtheilung der Amblyoponinae darf nach meiner Ansicht nur als Tribus in der Subfamilie der Ponerinen beibehalten werden."

⁵ *Loc. cit.*, p. 694, footnote.

male genitalia of *Stigmatomma serratum* Hald. are constructed "soweit es ohne Zergliederung zu sehen ist, ganz nach dem gewöhnlichen Ponerinentypus."

It would seem, therefore, that there are no very cogent reasons for adopting the subfamily Amblyoponinae, so far as characters drawn from the adult structure are concerned. The habits of *Stigmatomma*, as I have shown, are essentially the same as those of the Ponerinae, so that there exist no ecological grounds for accepting Forel's suggestion. The larva, however, seems to me to show very clearly that there is a greater gap between the Amblyoponii as a tribe of Ponerinae, and the tribes Ponerii and Odontomachii, than between the two last-mentioned groups. It must be remembered, however, that the larvae of two tribes of Ponerinae, the Australian Myrmecii and the cosmopolitan Ectatommi, have not been described, and that when these are known the striking differences between the Amblyoponii and the Ponerii may be reconciled. If, as Emery suggests,¹ the Myrmicinae are descended from the Ponerinae, it is obvious from a study of the larvae that the former subfamily must have come from forms with larvae like the Amblyoponii, a group which in other respects also is generally regarded as very primitive.² For the present I can see no reasons for altering the excellent classification outlined by Emery in his *Dorylus* paper.

There still exists an ecological (or biological, in the German sense) connection between the Ponerinae and the Myrmicinae, as I have lately ascertained. Since describing the peculiar method employed by the Texan Ponerinae in feeding their larvae, I have found that one of our New England Myrmecine ants, *Stenamma* (*Aphaenogaster*) *fulvum* Rog., subsp. *aquia* Buckl., var. *piccum* Emery — an ant very common under stones and in rotten logs along the edges of woods — has essentially the same method of feeding its young! My attention was first

¹ *Loc. cit.*, p. 773. "Dagegen liefern die Ponerinen offenbar die Wurzel, aus welcher die übrigen Unterfamilien der Ameisen (*i.e.*, after excluding the very primitive Dorylinae!) d. h. die Myrmecinen, Dolichoderinen und Camponotinen entsprossen sind."

² Emery, *loc. cit.*, p. 774, suggests that the Myrmicinae may be related to the Ectatommi, while the Dolichoderinae are more closely allied to the Ponerii.

called to this fact in an artificial nest belonging to Miss Adele M. Fielde, at Woods Holl, Mass. One afternoon Miss Fielde left a lot of queen pupae and larvae of *Crematogaster lineolata* within reach of the *Stenamma* colony. By the following morning the *Stenamm*as had carried these into their nest, cut off their heads and abdomens, and had distributed the pieces freely among the larvae, which could be seen singly and in groups of from two to five eagerly feeding on the juices in the same manner as Ponerine larvae. Thinking that this might be a very exceptional action, due to the confinement of the colony, I opened numerous nests in the woods during the month of August, while the ants were rearing their second brood. In nearly every one of these nests I found one or more larvae feeding on substances left among them by the workers. In one nest three larvae were feeding on a small Geometrid caterpillar; in another several had their heads and necks inserted into the thoraces of some small Carabid beetles that had been decapitated by the ants; in still another nest several larvae were devouring the pulp of a blackberry, etc.¹

Since making the above observation I find that Janet has recorded some very similar facts.² He saw several large larvae of *Lasius mixtus* and *L. flavus* sucking the juices from the cadavers of small larvae of their own species. Another more detailed observation I quote *in extenso*: "C'est dans un nid artificiel de *Tetramorium caespitum* que j'ai pu faire, à ce sujet, l'observation la plus précise. Au moment où les Fourmis récoltées venaient de terminer leur emménagement, j'ai vu, de la façon la plus nette, une larve d'ouvrière sucer une petite larve jaune de Coléoptère. La larve de *Tetramorium* n'était pas très éloignée d'avoir atteint sa taille définitive. Elle était suspendue par ses poils d'accrochage contre la paroi du nid, immédiatement sous le plafond en verre. Elle était placée horizontalement, le dos en haut, mais un peu de côté.

¹ *Stenamma fulvum* also brings into its chambers numerous seeds and the corollas of small white flowers. I mention this fact because it shows that this ant, which is normally carnivorous, nevertheless has proclivities that ally it by instinct as well as by structure to the harvesting ants of the subgenus *Messor*.

² *Le Lasius mixtus*, etc., *loc. cit.*, pp. 10-12.

Au-dessous d'elle, placée tête-bêche, parallèlement à son corps et soutenue en partie par les poils d'accrochage de l'abdomen du *Tetramorium*, se trouvait la petite larve jaune vermiforme, ayant $\frac{1}{2}$ millimètre de diamètre et 2 millimètres $\frac{1}{2}$ de longueur. Cette larve jaune avait certainement été placée là par une ouvrière, car pendant l'emménagement, j'en avais vu une qui introduisait une larve semblable dans le nid. La larve de *Tetramorium* avait sa tête infléchie et appliquée contre la larve jaune. Elle laissait voir, très nettement, sa bouche et ses pièces buccales. Grâce à ces circonstances exceptionnellement favorables j'ai pu examiner, avec une forte loupe, ce qui s'est passé, et cela pendant plus d'un quart d'heure. J'ai d'abord constaté le mouvement incessant de la bouche et vu nettement l'absorption du liquide transparent qui sortait de la plaie. Libre dans sa partie moyenne, la petite larve jaune était soutenue dans sa région céphalique par les poils d'accrochage de l'abdomen du *Tetramorium*. Ce dernier maintenait, au moyen de ses mandibules crochues, l'extrémité anale de sa proie, et cette extrémité était animée d'un mouvement rythmé de balancement résultant des mouvements de succion. Pendant ce repas, et sans que la larve du *Tetramorium* parut en être dérangée, une ouvrière est venue la lécher. Cette ouvrière est allée, ensuite, dégorger de la nourriture contre la bouche d'une larve voisine. Au bout d'un quart d'heure j'ai dû interrompre l'observation parce qu'une ouvrière est venue, malencontreusement, intercaler une nymphe entre la larve et le verre. J'ai alors pris la larve avec un pinceau et une petite cuiller et j'ai constaté qu'elle avait ramené sa bouche contre son corps, et que le repas était interrompu. Quant à la petite larve jaune dont j'avais vu le corps bien gonflé au commencement de l'observation, elle était, maintenant, surtout dans la région sucée, flasque et en partie vidée." Janet concludes his observations on this subject with the words: "Cette observation, venant confirmer celles, un peu moins précises, que j'ai faites chez d'autres espèces, ne me laisse plus aucun doute sur la faculté que les larves de Fourmis possèdent de sucer directement les liquides contenus dans le corps de larves, probablement blessées au préalable, qui les ouvrières déposent auprès

d'elles. Toutefois, cette manière de prendre la nourriture doit être considérée comme tout à fait exceptionnelle."

The fact that this peculiar method of feeding the larvae is the only method adopted by the Ponerinae, and, I believe, also by the Myrmicine *Stenamma fulvum*, and that it occurs as an exception in such highly specialized ants as *Lasius* and *Tetramorium*, is of considerable interest from the standpoint of the phylogeny of instincts. It not only spans a gap between the generalized Ponerinae and the more specialized Formicinae, but it would seem to indicate that the method of feeding the larvae by regurgitation was grafted on to this original method in the more recent ants, possibly in connection with their habit of feeding one another by regurgitation in their adult conditions.¹

In conclusion, the various peculiarities which indicate that the Ponerinae are a very primitive and generalized subfamily of ants may be enumerated²:

1. The colonies of the Ponerinae consist of a comparatively small number of ants, like the incipient colonies of the Myrmicinae, Dolichoderinae, and Formicinae.
2. These small colonies appear to be annual growths, formed by swarming, as in the bees, and not by single fertilized female ants unaccompanied by workers, as in the above-mentioned subfamilies.
3. Two or more colonies of Ponerinae of the same species can be fused to form a larger colony without much difficulty. This is not so easily accomplished with many species of the more specialized ants.

¹ Since completing my manuscript I find that one of our Texan Myrmicine ants (*Pheidole* sp. near *P. fabricator* Smith) resembles *Stenamma fulvum* in its manner of feeding the larvae. September 27 I opened a small nest of the *Pheidole* near Austin and found dozens of larvae feeding on fragments of different insects collected and comminuted by the workers. This is of interest because the *Pheidole*, like its congeners, is a harvesting ant, storing the large flat chambers of its nest with many seeds.

² While I have assumed in this and in my former paper that the Ponerinae may be regarded as the group from which the Myrmicinae, Dolichoderinae, and Formicinae (Camponotinae) have developed, I am quite of Emery's opinion that the existing Ponerinae are not ancestral forms. Emery calls attention to the fact that the palpi are aborted in all the tribes of Ponerinae except the Myrmecii, whereas many of the genera of higher ants have retained the undiminished number of joints in these organs.

4. The architecture of the Ponerinae is of a primitive character, consisting of a few irregular and unfinished galleries and chambers. The latter are not even formed by all the species.

5. The queen and worker differ but little in size and structure.

6. Ergatoid females, or forms intermediate between the queens and workers, are of normal and comparatively frequent occurrence in some species.

7. The habits of the queen and worker are very similar. The female is not an individual on whom special attention is bestowed by the workers.

8. The workers show no tendency to differentiate into major and minor castes.

9. The Ponerinae are carnivorous and live by hunting (in contrast with the various harvesting, fungus-growing, honey-collecting, and aphidicolous members of the more specialized subfamilies).

10. They do not feed one another by regurgitation.

11. The larvae are not fed by regurgitation, but are given pieces of insects from which they suck the juices.

12. The cocoon is retained as a pupal envelope throughout the group. (The Ecitonii among the Dorylinae have lost this envelope, although the Dorylii still retain it; it is lost in the Myrmicinae, and is apparently in the process of disappearing among the Formicinae.)

13. In at least one genus (*Stigmatomma*) the callows are able to escape from their cocoons without the assistance of the workers.

14. The callows of the Ponerinae are more mature on leaving the cocoon than the newly hatched Formicinae.

COLEBROOK, CONN., Sept. 5, 1900.

FUSION OF FILAMENTS IN THE LAMELLIBRANCH GILL.

EDWARD L. RICE.

It is far from the purpose of this paper to enter fully into the discussion of the morphology of the lamellibranch gill; but a brief preliminary statement of a general nature may serve to define the terminology employed, and to simplify the following more detailed description of a single point in the gill structure.

When the animal is oriented in the usual manner, with the hinge line upward, the gills hang down on either side, between the body and mantle, as shown in Fig. 1. Strictly considered, there is but one gill, or one ctenidium, on each side of the body. This ctenidium consists, fundamentally, of a slight longitudinal ridge along the side of the body, the gill axis (Fig. 1, *a*), and a double row of ciliated filaments, which extend downward into the mantle cavity of the animal and are then reflected upward. In common language each of these rows of filaments, or the structure arising therefrom, is termed a gill; and I shall retain this convenient, though morphologically indefensible term, and designate that half of the ctenidium next the body as the inner gill (Fig. 1, *b*), and that half next the mantle as the outer gill (Fig. 1, *c*).

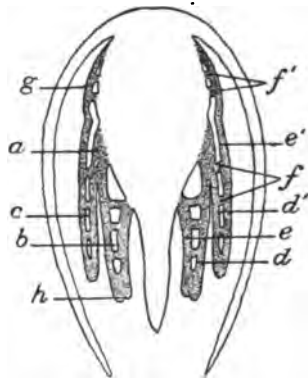


FIG. 1. — Diagrammatic frontal section of lamellibranch. Shell omitted; gills shaded. *a*, gill axis; *b*, inner gill; *c*, outer gill; *d* and *d'*, descending lamellae of inner and outer gills; *e* and *e'*, ascending lamellae of inner and outer gills; *f* and *f'*, interlamellar connections of gill and dorsal appendage; *g*, dorsal appendage of outer gill.

In some few forms the adjacent gill filaments remain entirely free from one another, *e.g.*, *Anomia*; in other forms, as *Mytilus* and *Pecten*, there is a union by means of tufts of very long interlocking cilia — ciliated discs. In contrast to these two types,

which may be grouped together as filamentous gills, we find in the great majority of lamellibranchs a more or less strong development of vascular connections between the filaments, binding them together to form the lamellae from which the class takes its name. Such gills may be termed lamellar.

As each filament consists of two limbs, a descending and an ascending, so each gill is composed of two lamellae, correspondingly designated as descending (Fig. 1, *d* and *d'*) and ascending (Fig. 1, *e* and *e'*). These lamellae are usually connected with one another by more or less complicated interlamellar connections (Fig. 1, *f*).

The ascending lamella of the inner gill may remain free, or its upper margin may fuse with the body and, behind the body, with the corresponding part of the gill of the other side. The ascending lamella of the outer gill may lie free in the mantle cavity or may be attached to the mantle on a level with the gill axis; or it may be continued dorsally above this line, forming a dorsal appendage (Fig. 1, *g*), which is finally attached to the body, or rather to the fusion line of mantle and body. Thus the dorsal appendage consists of the ascending lamella alone, and may show a structure decidedly different from that of the gill proper. Connections similar to the interlamellar connections of the gill attach the dorsal appendage to the body wall (Fig. 1, *f'*).

The lower free margin of some gills is smoothly rounded off; in other cases the border is deeply notched by a groove running from end to end of the gill. This may be called the marginal groove. In the diagram the groove is seen in cross-section in the inner gill (Fig. 1, *h*), while the outer gill presents a smooth margin with no sign of a furrow. Exactly these conditions are found in a large number of lamellibranchs, *e.g.*, *Astarte*, *Dreissenia*, *Cardium*, *Psammobia*. It may be suggested in passing that the somewhat unintelligible distinction of "double gills" and "single gills," in the classical paper by Williams,¹ may perhaps be based on the presence or absence of this marginal groove.

¹ Williams, T., "Respiratory Organs of Invertebrates," *Annals and Magazine of Natural History*. Vol. xiv. 1854.

The surface of the gill lamellae may show either of two types. It may be smooth, all the filaments lying in one plane; or, with the requirement of greater respiratory surface, the lamellae may be folded parallel to the filaments. The two lamellae of one gill are always folded symmetrically, so that the section of the gill perpendicular to the filaments assumes an outline reminding one, in its extreme form, of a string of wooden button molds. Both filamentous and lamellar gills are subject to this folding.

The filaments occupying the bottom of the reëntrant folds, the limiting filaments, are usually somewhat larger than the intermediate filaments, and are often very much modified in form as well as size. These filaments may be easily traced through the whole height of the gill and afford a series of fixed points, aiding materially in the study of the folded type of gill by means of sections.

From the above résumé it is evident that fusion or concretion of parts has long been recognized in the lamellibranch gill. Even in simple filamentous gills (*Mytilus*, etc.) the tips of the filaments fuse to form a continuous band along the margin of the ascending lamella. In more complex forms it is a process of fusion which transforms the rows of originally distinct filaments into the characteristic lamellae. The fusion of the ascending lamellae with the mantle and body is also perfectly familiar. But the particular type of fusion described below appears to have escaped mention in the somewhat extensive literature upon the lamellibranch gill.

Even a hasty study of serial sections of the strongly folded gill of *Cardium edule* or *Batissa tenebrosa* shows the somewhat surprising fact that each fold contains a far larger number of filaments in the upper portion of the gill than in the neighborhood of the free margin, this being equally true of ascending and descending lamellae. (The extreme case was noted in *Cardium*, where the number of filaments in one fold increased from eight to thirty.) Yet the limiting filaments are continuous throughout, showing that there is no reduction in the number of folds correlated with the increase in the number of filaments in each fold. These observations appear at first sight irreconcilable with

the theory of gill development advanced by Lacaze-Duthiers¹ and universally accepted, according to which the ascending limbs of the filaments are essentially the reflected tips of the originally simple straight filaments. The larger number of filaments

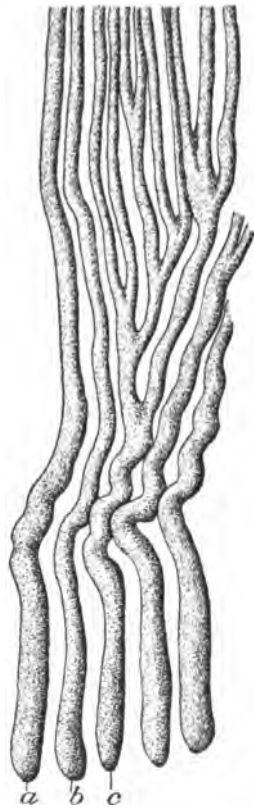


FIG. 2. — Surface view of group of filaments of inner gill of *Batissa tenebrosa*. $\times 60$. Lower edge of figure represents the free margin of gill. Cilia and complex interfilamentary connections omitted for the sake of clearness. *a* and *b*, simple filaments; *c*, compound filament, formed by fusion of nine simple filaments.

in the upper parts of the descending lamellae could be explained by the assumption of the presence of a large number of abortive filaments. But this explanation, unsatisfactory even for the descending lamellae, cannot apply to the ascending lamellae.

A more detailed study of serial sections, and, better, of microscopic dissections in which the lamellae are separated and the folds spread out as smooth as possible, shows clearly that the filaments of the upper portion gradually meet and fuse as the free margin of the gill is approached. This phenomenon is illustrated by Figs. 2 and 3. In the former we have a surface view of a small portion of the inner gill of *Batissa tenebrosa*, extending upward from the free edge. While the two filaments at the left (Fig. 2, *a* and *b*) are simple throughout, the next filament (Fig. 2, *c*), indistinguishable from the others at its lower end, is really formed by the fusion of nine filaments.

In this species such groups of fusing filaments are found principally in the projecting folds of the gill, very seldom in the reëtrant folds. In *Cardium edule*, on the other hand, the fusion

is more marked in the reëtrant than in the projecting folds.

¹ Lacaze-Duthiers, H. de, "Mémoire sur le développement des branchies des Mollusques Acéphales Lamellibranches," *Annales des Sciences Naturelles*. Zoologie Sér. iv, Tome v. 1856.

Fig. 3 represents diagrammatically the surface of a single fold of this species, each filament being represented by a simple line. At the gill margin (lower side of diagram) there are eight filaments, including the limiting filaments (Fig. 3, *a* and *h*); in the upper part of the gill (and diagram) there are thirty. Of these thirty filaments, twenty-one unite to form the two limiting filaments (Fig. 3, *a* and *h*) and the two adjacent intermediate filaments (Fig. 3, *b* and *g*), which occupy the re-entrant folds. The remaining four intermediate filaments (Fig. 3, *c*, *d*, *e*, and *f*), those of the projecting fold, are formed by the union of only nine filaments. The irregularity of the fusion is also well shown by *a*

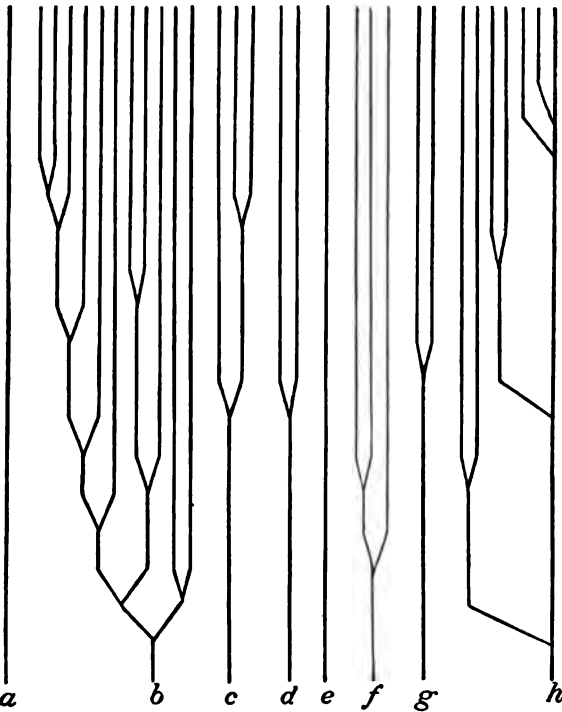


FIG. 3.—Diagrammatic view of surface of fold of inner gill of *Cardium edule*, illustrating fusion of filaments. Filaments represented by lines. Lower edge of diagram represents free margin of gill. *a* and *h*, limiting filaments; *b-g*, intermediate filaments.

this diagram. Note especially that one limiting filament is entirely simple, while the other is the resultant of the fusion of seven simple filaments.

The fusion is usually almost exclusively limited to a somewhat narrow zone in the near vicinity of the free margin of the gill. In this zone may also be noted a gradual reduction in the folding of the gill as the margin is neared. In those lamelli-branches whose outer gill is provided with a dorsal appendage, there is a second zone of fusion along the transition line from

the gill proper to the appendage. This line again marks a partial or total obliteration of the folding of the gill — a point of importance in the consideration of the meaning and cause of the fusion of the filaments.

The term "fusion of filaments" has been employed repeatedly; but the question arises whether the phenomenon under discussion is really a fusion of a large number of once distinct filaments or a branching of a relatively small number of original filaments; whether, in other words, the primary filaments are represented by the maximum or minimum number. The answer is not far to seek. At the free margin of the gill the continuity of the filaments may be readily traced from one lamella to the other, and the number of filaments in the two lamellae within any given fold must necessarily be equal. A little higher, in the zone of fusion, these numbers may become very unequal. For example, in a section of the gill of *Batissa* seven filaments of one lamella were observed to correspond to twelve in the other. Neither of these facts, however, offers conclusive evidence; both may be explained as well on the supposition of branching as on that of fusion. But as the serial sections are followed a little higher in the gill, the inequality in the number of filaments, which has increased from zero at the margin to a maximum in the zone of fusion, begins to fall off again more or less rapidly, and finally reduces itself once more to zero. This equality in the number of filaments in the upper parts of the gill can be explained only on the supposition that the phenomenon before us is a fusion, not a branching, of the gill filaments. It is altogether too improbable that the independent branching of the filaments in the two lamellae would lead to the same number in the two cases.

Granted that the phenomenon is a fusion, what is its meaning? Is it of systematic importance? My observations are not complete enough to permit a categorical answer; but I am strongly inclined to the belief that the fusion carries no more of weight from the systematic standpoint than does the folding of the gill.

Among smooth gills no fusion was observed, although a considerable number of forms were studied. Especial attention was devoted to *Tellina* and *Scrobicularia*, in which, for reasons

detailed in an earlier paper,¹ I consider the simplicity and smooth surface of the gill to be secondary characters — a retrograde development from the folded gill type of the Veneridae. Fusion was also never observed in the filamentous type of gill.

The results of my own observations upon the folded lamellar gills may be tabulated as follows :

1. Fusion strongly developed: *Cardium edule* Lin., *Chama pellucida* Brod., *Batissa tenebrosa* Hinds, *Psammobia vespertina* Lin., *Donax serra* Chemn.
2. Fusion moderately developed: *Venus verrucosa* Lin., *Cyprina islandica* Lin., in latter strongly developed on transition line from outer gill to appendage.
3. Fusion very slightly developed: *Eusatella americana* Verrill, *Mya arenaria* Lin., *Donax politus* Poli, in latter perhaps accidental.
4. No fusion observed: *Cytherea chione* Lin., *Donax trunculus* Lin., *Ostrea virginiana* Lister, *Thracia papyracea* Poli.

Lima and *Ostrea* should probably be added to the list of forms in which fusion occurs, though on somewhat doubtful evidence, which will be mentioned later.

Thus we find this fusion of filaments similarly developed in widely separated forms, belonging to very diverse groups; on the other hand, within the single genus *Donax* we find all grades of fusion, as we also find all grades of folding.

This parallelism of folding and fusion in the genus *Donax* appears to me to furnish, in a certain sense, an epitome of the whole matter, for I consider the fusion of the filaments to be a mechanical correlative of the folding of the lamellae. The process may be pictured in something this way. The folding of the lamellae is gradually developed in the young lamellibranch as the increasing number of filaments becomes too great to lie in one plane. But at the free margin the filaments are bound somewhat firmly together and the folding is somewhat reduced. This leads to a crowding together of the filaments at this point and eventually to an organic fusion of the same. How great the crowding must really be in the strongly folded forms may be inferred from the fact that the upper part of the inner gill of

¹ "Die systematische Verwertbarkeit der Kiemen bei den Lamellibranchiaten," *Jenaische Zeitschrift für Naturwissenschaft*. Bd. xxxi. 1897.

Cardium edule would measure, if flattened out, fully seven times the length of the free margin of the same gill. The absence of fusion in filamentous gills, even where the folding is extreme, as in *Pecten*, may be easily explained on the ground of the looser structure of the gill and the possibility of a displacement of the filaments, with consequent relief of pressure.

It is an interesting point in this connection that no fusion was observed in the outer gill of either *Psammobia* or *Cardium*, although it is conspicuous in the inner gill. In both cases the inner gill is provided with a deep marginal groove, while the outer gill shows no sign of this structure. The presence of this marginal groove is the mechanical equivalent of a shortening (slight to be sure) of the margin of the gill, the bottom of this groove being almost perfectly straight, and therefore slightly shorter than the somewhat scalloped margin of the folded gills in which no groove is present. Hence we find in this characteristic a cause of increased crowding and increased fusion.

For a very short distance above each point of fusion one observes a slight modification of the epithelium of those sides of the fusing filaments which are turned toward each other. The nuclei are somewhat larger and more closely crowded, as shown diagrammatically in Fig. 5. Aside from this, no histological distinction can be drawn between the simple original filaments of the upper portion of the gill and the compound filaments of the lower margin. Even in the matter of size there is no noticeable difference except in the immediate vicinity of the fusion, where the compound filament is considerably enlarged. This statement is made after comparison of a large number of preparations, and seems to be the rule, although there are considerable individual variations in different specimens. The apparent larger size of the compound filaments in Fig. 2 is to be explained in part on the ground of such individual variations. It is due in larger degree, however, to slight differences in the position of the filaments, which are elliptical in cross-section, and appear of different size according as one or another side is presented to view. It should also be noted that the filaments are somewhat enlarged at the extreme margin of the gill, and that these enlarged tips are shown in the figure.

As regards the size and finer structure of the filaments, the evidence of sections is more reliable than that of surface preparations; and in Figs. 4-8 are represented a series of sections through a group of filaments of the inner gill of *Cardium edule*. The same filaments are shown in all the figures, although the very complex interfilamentary and interlamellar connections, differently cut in the different sections, cause a considerable variety of aspect. These connections may be disregarded in the present discussion. In Fig. 4, which represents the uppermost section, six filaments are shown, all practically alike. In



FIG. 4.



FIG. 5.



FIG. 6.

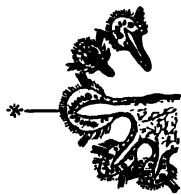


FIG. 7.

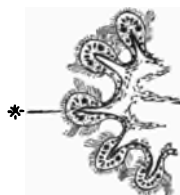


FIG. 8.

FIGS. 4-8. — Series of sections through group of filaments of inner gill of *Cardium edule*. $\times 95$. Cilia and nuclei somewhat diagrammatic. Fig. 4 is uppermost section. Asterisk marks two simple filaments fusing to one compound.

Fig. 5 two of these, marked with an asterisk in all the figures, show a slight modification of the epithelium, as described above. This section is immediately above the point of fusion of the two filaments. Fig. 6 shows these filaments so closely approximated that they may almost be described as forming a single deeply grooved filament, a condition better shown by other preparations. In Fig. 7 the fusion is complete, but the compound filament is still considerably enlarged; while in Fig. 8 the compound filament has regained its normal size and form, and the transition is complete.

Strangely, this very conspicuous fusion of the filaments appears to have received little or no notice. I have been able to find no reference whatever in the text of any articles at my

disposal. Only in the figures is a possible hint of its observation. Thus in the exquisite work by Deshayes¹ on the Mollusca of Algiers there is a small figure of the gill of *Pholas*, in which two incomplete filaments are interpolated among those which extend through the whole height of the gill. In the one the free end is turned upward; in the other, downward. Do these represent two filaments fusing with their neighbors? If so, the difference in direction in the two cases clearly distinguishes the phenomenon from that described above. Moreover, the author's description of the gill as entirely smooth makes a fusion *a priori* improbable. An inaccuracy in the drawing, which is very small, is the simpler explanation.

In a considerable number of more recent figures a single fold of the gill is represented in section as containing an unequal number of filaments in the two lamellae, thus in *Cardium* (van Haren),² *Lima* (Pelseneer),³ *Donax trunculus* (Sluiter),⁴ and *Ostrea* (Kellogg).⁵ It will be noted that in the last two cases the figures cited do not accord with my own observations. But my purely negative verdict of "not observed" contains no direct contradiction of the affirmative statements of these authors. While I am convinced that my own preparations show no evidence of a fusion of filaments in *Ostrea* and *Donax trunculus*, I consider it probable that the figures of Kellogg and Sluiter, as well as those of van Haren and Pelseneer, point to a greater or less development of the phenomenon described. Unfortunately no certain conclusions can be reached in the absence of corroboration in the accompanying text.

¹ Deshayes, G. P., "Exploration scientifique de l'Algérie," *Zoologie*. Tome i, No. 4. Paris, 1849.

² Haren-Noman, D. van, "Die Lamellibranchiaten, gesammelt während der Fahrten des Willem Barents, 1878 and 1879," *Niederländisches Archiv für Zoologie*. Supplementband I, 1881-82.

³ Pelseneer, P., "Contribution à l'étude des Lamellibranches," *Archives de Biologie*. Tome xi. 1891.

⁴ Sluiter, C. P., "Beiträge zur Kenntniss des Baues der Keimen bei den Lamellibranchiaten," *Niederländisches Archiv für Zoologie*. Bd. iv. 1878.

⁵ Kellogg, J. L., "A Contribution to our Knowledge of the Morphology of the Lamellibranchiate Mollusks," *Bulletin of the U. S. Fish Commission*. Vol. x. 1890.

PORTABLE ANT NESTS.

ADELE M. FIELDE.

IN order to keep ants under continued observation, and at the same time to change occasionally the domicile of the observer, it is necessary to have portable nests. The transportation of either the Lubbock or the Janet nest is made inconvenient by the water used for the isolation of the ants in the former, and by the considerable weight of the latter.

Six colonies can be successfully carried on journeys of several hours or days by the use of a case made of half-inch pine boards and dovetailed at the corners, with a door hinged upon its lower side and held shut by two buttons at its upper edge. The case measures on the inside $16\frac{3}{4}$ inches in length, $6\frac{1}{4}$ inches in horizontal depth, and $4\frac{3}{4}$ inches in height. Three shelves, each one-fourth of an inch thick, are mortised into the ends of the case, making four compartments, each one inch high. Several holes are bored in the door of the case and in the side opposite the door, to admit fresh air. The case is carried by a leather handle fastened lengthwise to its top, and it is just filled by the six nests hereinafter described.

Of the six nests, there are two of each pattern, A, B, and C. An A and a B nest together fill a shelf, while a C nest fills a shelf alone.

The nests are all built of clear glass, and their parts are readily cut by any glazier. The joinings are made with Le Page's liquid glue, which must thoroughly dry before the nests are used. The cost of material and the amount of labor required in making these nests are much less than for either the Lubbock or the Janet pattern.

The floors are panes of "double thick" glass, under which is laid a thick sheet of white blotting paper of the same size as the glass, to give opacity, elasticity, and a background against which the ants are easily seen. The blotting paper should not

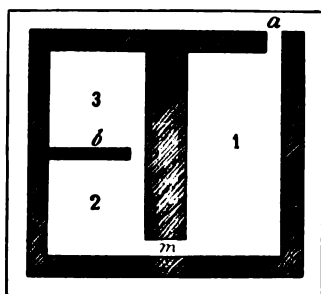
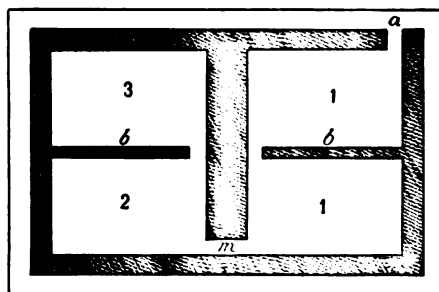
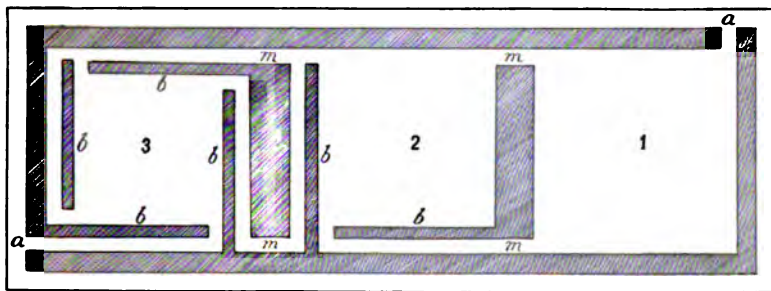
be fastened to the glass, as the latter must sometimes be raised in order to get a view of the ants from below.

The walls stand half an inch from the edges of the base-pane, for the greater security of the superstructure when the nest is being lifted. The walls are built of strips of "double thick" glass, half an inch wide, two horizontal layers in every wall giving a perfectly level top. Apertures three-eighths of an inch wide are left in the walls at points marked *a* on the plans. These apertures admit the end of a three-eighths-inch glass tube, which may be used as a safe bridge between nests when ants are to be made to pass from one nest into another. When not in use, the apertures are closed with plugs of cotton. The outside of the walls, after thorough drying, are painted black, to secure within the nest the darkness in which ants like to keep their young.

The main partitions in the ant-dwelling take part with the walls in the support of separate roof-panes, and they are therefore twice as wide as are the walls. Passageways (marked *m* on the plans) permit the ants to go from one room to another, as is necessary when a room is to be cleaned. The admission of strong light will insure the removal of the ant family to the darkened chamber adjoining. The passageways may be from one-fourth to one-half inch in width, and they should be covered with very thin glass, well glued on.

The tops of the walls and main partitions are exactly covered with coarse Turkish toweling, cut in strips twice as wide as the base on which it lies, and doubled so that its raw edges meet in the middle of the glass it doubly covers. The under layer of the toweling is glued to the glass, and it performs the double office of keeping the ants within the nest and of admitting sufficient fresh air for their breathing. Just over the passageways the toweling is left free, so that it may there be lifted for observation of what is within. After the toweling is laid on, the rooms have a uniform depth of less than half an inch, and a hand lens can be focused upon any of their inmates. The toweling being elastic and level, the roof-panes, of thin, clear glass, lie closely upon it. They are not fastened down.

The roof-panes reach the center of the main partitions and

A. $6\frac{1}{2} \times 6$ in. ($16\frac{1}{2} \times 15\frac{1}{2}$ C.)B. 10×6 in. ($25\frac{1}{2} \times 15\frac{1}{2}$ C.)C. $16\frac{1}{2} \times 6$ in. ($42 \times 15\frac{1}{2}$ C.)

1. Food-room.

a. Entrance.

2. Nursery.

b. Screen.

3. Sponge-room.

m. Passage.

project one-fourth of an inch beyond the walls. All the rooms except the food-room have an outer roofing of thick dark blotting paper, which should be lifted only when actual study of the ants is proceeding.

In the rooms numbered 1 the ants have, as in the Janet nests, a chance to range in the light and to seek food, of which it is well to put in the smallest sufficient quantity and several kinds. If the ants are made to move into the darkened food-room, leaving the other rooms free for cleaning, the passageways (*m*) may, during the cleaning, be plugged with cotton.

In room 3 a soft, fine sponge, clean and wet, and less than one-fourth of an inch thick, should nearly cover the floor, leaving a passage all around it next the walls. This furnishes drink to the ants and moisture to the air of their dwelling. If the ant young are in the egg or the larval stage, or if the temperature is high, the floor of room 2 should likewise be covered with wet sponge; but if the young are in cocoons, or if the temperature is very low, then room 2 should have a layer of wadding instead of sponge. The ants generally choose damp places for the eggs and larvae, and dry places for the cocoons or pupae.

The screens (marked *b*) are substitutes for the ant-runs used in the ground, and they gratify the disposition of the ant to keep close to cover in going about in the nest. They are made in the same way as are the walls, but are only one-fourth of an inch thick, and are not topped with toweling.

The A nest, with base $6\frac{1}{2} \times 6$ inches, is designed for a colony of very small ants, or for a few large ants. The B nest, with base 10×6 inches, affords a home for a somewhat larger family. The C nest, $16\frac{1}{2} \times 6$ inches, can be used for a multiplying and dividing colony, or for observing the activities of restless species. The ants should never be greatly crowded in their habitation.

The ants in my nests appear sleek and healthy. I have found these nests easier than others to keep free from the molds that grow from particles of food that the ants convey from the food-room to every other part of their nest. These nests also lend themselves readily to experimental uses in studying the instincts of their occupants.

When the nests are to be carried on a journey the roofs are securely fastened down by sewing narrow strips of cheese cloth around the nest in such a way as to prevent the slipping of the roof-pane. The fastenings must not exclude fresh air. Having fastened the roof-panes each in place, the nests are put upon the appropriate shelves in the case, where they may be further secured by bits of wadding above the roof-panes and at the ends of the shelves.

The weight of my case, with its six enclosed nests packed for travel, is less than fifteen pounds. The strong local attachments of the ants are undisturbed by their so journeying, and at the end of the journey the study of their life processes may be speedily resumed.

MARINE BIOLOGICAL LABORATORY,
WOODS HOLL, September, 1900.

BIOLOGICAL BULLETIN.

THE OESOPHAGEAL GLANDS OF URODELA.

R. R. BENSLEY.

FOR a long time the only known instance of glands occurring in the oesophagus of an Amphibian was the familiar pepsin-producing glands of the frog's oesophagus, discovered as early as 1838 by Bischoff (1). In 1853 Leydig (8) described the occurrence of saccular glands in the oesophagus of *Proteus anguineus*, and, more recently, similar glands have been discovered by Kingsbury (5) in the oesophagus of *Necturus maculatus*.

In no other Batrachian has investigation revealed the existence of glands in the oesophagus, unless, indeed, as Klein (6) suggests, the highly branched glands found at the junction of oesophagus and stomach in Triton, and termed by Langley (7) the anterior oxyntic glands, are such.

Naturally, considerable interest has been evinced in the question of the homology of these glands one with another, and with those of the higher vertebrate classes.

In order that the problem to be solved may be clearly understood, it may be as well to recapitulate briefly the facts as they appear in the forms so far investigated.

In the frog, leaving out of consideration the pyloric glands, there are two kinds of glands occurring in the foregut. The oesophageal glands occur under a ciliated epithelium, and are large compound glands, consisting each of a number of short tubular acini lined by pepsin-secreting cells, opening into a common duct lined by transparent mucous cells. As we pass

down the oesophagus we find that, at the point where the ciliated epithelium is succeeded by the ordinary mucigenous epithelium of the stomach, there is a gradual transition from the compound oesophageal glands to the more simple tubular glands of the stomach, the second type. The secreting cells of the two kinds of gland differ markedly from one another. Those of the gastric glands contain few zymogen granules of small size, while those of the oesophageal glands are more or less filled with very large granules, and the cells themselves are larger. Further, the oesophageal glands yield an alkaline secretion, the gastric glands an acid secretion.

In Triton, again, there are two types of gland. At the junction of oesophagus and stomach occur the anterior oxyntic glands of Langley. The difference between these glands and the other gastric glands (posterior oxyntic glands of Langley) is not so marked. The former are more highly branched and are separated from one another by a larger amount of connective tissue, but the differences in the size of the granules and in the nature of the secretion, so conspicuous in the case of the frog, are absent.

In Proteus a new structure makes its appearance in the shape of isolated sac-like glands occurring in the oesophagus. These have been fully investigated by Oppel (11), who describes them as follows: "Die Drüsen des Oesophagus haben eine rundliche Form. Sie bestehen aus einem grossen Acinus. Die Drüsen sind zusammengesetzt aus einem Ausführungsgang und dem secernierenden Theil. Ich spreche von einem Ausführungsgang, da sich die Zellen desselben von denen der Schleimhautoberfläche unterscheiden. Der Ausführungsgang besteht aus Zellen von annähernd cylindrischer Form, und zwar ist die Grenze zwischen conischem und cylindrischem Epithel stets eine scharfe. Eine besondere Eigenthümlichkeit liegt in der Uebergangsstelle von diesen cylindrischen Zellen des Ausführungsganges zu den secernierenden Zellen. Dieselbe liegt nämlich nicht an der Stelle, an welcher die Erweiterung des engen Ganges zum Acinus stattfindet, sondern die Cylinderzellen gehen noch ein Stück weit in den Acinus hinein, um dann rasch zu den niedrigeren secernierenden Zellen abzufallen.

Diese Zellen zeigen in ihrem Protoplasma einen körnigen Bau, Körner, welche sich mit verschiedenen Farben, z. B. Eosin, S.-Fuchsin tingieren, mit Osmiumsäure bräunen" No glands resembling the anterior oxyntic glands of Triton are present in the adult, but he found in the young animal, at the junction of oesophagus and stomach, glands which are intermediate in nature between the oesophageal and gastric glands.

In *Necturus* there are, according to Kingsbury, three kinds of glands present. In the oesophagus are large saccular glands in most respects like those of *Proteus*, except that Kingsbury was unable, even after repeated trials, to demonstrate the presence of any granules capable of reducing osmic acid. At the junction of oesophagus and stomach are richly branched glands like the anterior oxyntic glands of Triton, and finally there are the ordinary gastric glands.

There are thus three types of gland occurring in the oesophagus of Batrachia, the relationship of which to one another, to the gastric glands, and to the oesophageal glands of higher vertebrates, is obscure. These are the compound pepsin-forming glands of the frog's oesophagus, the saccular glands of the oesophagus of *Proteus* and *Necturus*, and the anterior oxyntic glands of Triton and *Necturus*. It might be claimed for *a priori* reasons that no possible relationship could exist between the oesophageal glands and the gastric glands, but that position would necessitate a critical examination of the data on which this anatomical division of the foregut in the forms mentioned has been decided.

The writer found that *Amblystoma* combined, in a sense, the conditions found in *Proteus* and Triton, inasmuch as the glands in the larva resemble those of *Proteus*, the glands of the adult those of Triton. The present memoir is a brief account of the histogenesis of the glands in question.

Before passing on to a consideration of the histogenetic phenomena it is necessary to describe briefly the structure of the mucous membrane of the foregut in the adult animal.

The oesophagus is non-glandular, and is lined throughout by a ciliated epithelium, in which many goblet cells may be recognized. The ciliated epithelium is succeeded by the

ordinary cylindrical cells of the stomach at the point where the first glands appear, and the oesophagus expands suddenly into the stomach. The ciliated epithelium does not extend into the stomach.

The gastric zymogenic glands are of two kinds. The anterior oxyntic glands occupy the proximal portion of the mucous membrane and form a zone about 2 mm. in width around the

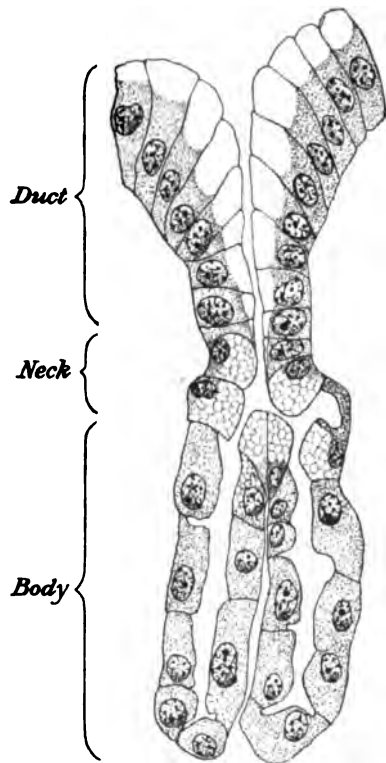


FIG. 1. — Gastric gland of *Amblystoma Jefferssonianum*. Zeiss apoch. 2 mm., ocular 2.

oesophageal orifice of the stomach. They are much shorter than the other gastric glands and, like the corresponding glands in Triton, are more highly branched, and contain more mucous neck cells. The rest of the stomach, with the exception of the posterior third in which the pyloric glands are found, is occupied by the usual tubular glands, consisting of a body composed of granular pepsin-forming cells, a neck composed of transparent mucous cells, and a duct composed of cells resembling the surface epithelium. These glands correspond in all respects to the excellent description given of the corresponding structures in Triton by Carlier (2). To make clear the terminology employed, a bi-tubular gland is represented in Fig. 1.

It was found necessary to resort to a new method of staining the zymogen granules, as the conventional method, by the employment of osmic acid, was not satisfactory when the cells contained brown pigment, or a great deal of prozymogen, which, as Langley (7) and Grützner (3) point out, also reduces the osmic acid, obscuring the granules if they be few in number or

of small size. For this purpose the writer employed Reinke's neutral gentian as follows: To a saturated solution of gentian violet in water a solution of orange G is added in excess. A brownish precipitate is formed which is very slightly soluble in water. This may be collected on a filter and washed until the wash water is only slightly tinged. The precipitate is then dissolved in alcohol. For use a sufficient quantity is added to twenty per cent alcohol to make a fluid of about the same color as a good solution of haemalum. Sections fastened to the slide are stained in this for twenty-four hours, all adherent stain is then removed by pressing down upon the sections several folds of filter paper, absolute alcohol added and quickly removed with the blotter, and finally oil of cloves added in which the differentiation of the stain takes place. As soon as the protoplasm of the epithelial cells appears orange, the extraction of the stain may be checked by washing in benzole, and the sections may then be mounted in the usual way in balsam. The zymogen granules are stained of an intense blue color, the nuclei blue, other portions of the cells orange. The stain is most successful after fixation in aqueous sublimate.

The earliest stages in the formation of the gastric glands are difficult to discern, owing to the great number of yolk spherules present which obscure the outlines of the cells. In a larva 11 mm. long the glands are already visible as tubular down-growths of the endoderm of the foregut. In this early larva two kinds of glands are already to be recognized, those occupying the anterior end just behind the tracheal groove, and those at the posterior end, where the stomach is as yet not clearly marked off from the general endoderm. The anterior glands are of a flask-like shape, and have a distinct lumen surrounded by a single layer of cells. Zymogen granules are not yet to be recognized, and the yolk spherules are so abundant that the outlines of the cells are not visible. In the lumen there may often be seen one or two cells, which have been, so to speak, squeezed out of the row of endoderm cells forming the gland. These cells do not take any part in the formation of the permanent histological elements, but may often be

recognized, even to a late stage of development, as disintegrating remains in these and the other gastric glands.

The posterior glands are simple tubes composed of a single layer of large yolk-filled cells surrounding a cleft-like lumen. Indeed, often it appears as if the endoderm of the foregut were in several layers without any differentiation into glands and epithelium. On careful inspection, however, it may be seen that the nuclei are arranged in an orderly fashion, as if surrounding the lumina of glands. Such an appearance is illustrated in Fig. 2. In this gland, in addition to the nuclei which are clearly arranged in a row around the lumen, two others may be seen which are nearer the center of the gland; these are the nuclei of cells which will later be found as disintegrating remains in the lumen.

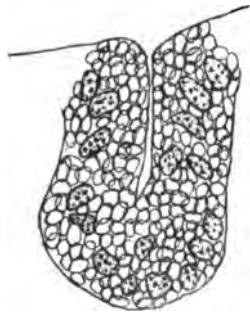


FIG. 2.—*Amblystoma* larva 11 mm. in length; gastric gland. Zeiss apoch. 2 mm., comp. ocular 2.

The flask-shaped glands do not, as one proceeds caudad, abruptly give place to the simple tubular glands, but there is a gradual transition.

In a larva 12 mm. in length, although the caudal portion of the stomach is still undifferentiated and the cells crowded with yolk, the yolk has sufficiently disappeared from the anterior portion to enable the shape of the glands and the cells composing them to be clearly determined. The anterior glands are now distinctly saccular, with a large lumen surrounded by a single layer of cells. The yolk spherules disappear from the cells of the glands somewhat more rapidly than from the surface epithelium, which as yet contains a considerable number. Notwithstanding the presence of the yolk, one can clearly distinguish, at this early stage, several kinds of cells, which can be readily referred to their analogues in the glands of the adult. The flask-shaped body of the gland (Fig. 3, *a*) is formed of a single layer of small cells, which vary from cubical to fusiform in shape and are usually convex towards the lumen. The protoplasm of these cells is granular and contains one or more yolk spherules. The nucleus is round or oval and rich

in chromatin. As the gland narrows into the duct (Fig. 3, *b*), these are replaced by two or three slightly larger cells, in each of which two zones may be recognized, an outer, wider, deeply staining zone containing the oval nucleus, and an inner one which stains but feebly. This inner zone exhibits a reticular structure due to the presence of a secreted substance, probably mucigen. In the more posterior tubular glands, likewise, two kinds of cells may be found, similar in all respects to those of the flask-shaped glands, granular cells occupying the body of the gland, mucigenous cells the neck. In sections stained in Reinke's neutral gentian it is found that already numerous zymogen granules are present in the deeply staining cells forming the body of the gland. In the pancreas, also, zymogen granules may be recognized long before the yolk granules have entirely disappeared from the cells.

The epithelium of the foregut in the region occupied by the flask-shaped glands is composed of two kinds of cells (Fig. 3, *c*), ciliated cells and cells the outer ends of which stain diffusely and intensely. These obviously represent the two characteristic elements of the future oesophageal epithelium, the ciliated and the goblet cells. Over the tubular glands farther back there is only one kind of cell in the epithelium, and this is without cilia.

Mitoses may be observed with equal frequency in all the various kinds of cell composing the epithelium and glands, and all are apparently equally capable of reproduction.

The important points to be learned from this stage are that the characteristic elements of the glands are differentiated very early, that no special groups of cells have, as yet, assumed the mitotic function to the exclusion of the others, and that a portion of the glandular foregut bears a ciliated epithelium.

In a larva 14 mm. in length the foregut has advanced to a considerable degree beyond the stage last described. It is now shaped like a letter U, with a long proximal and short

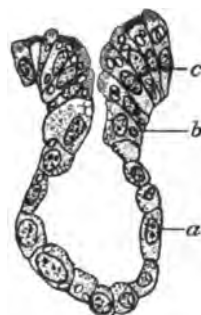


FIG. 3. — *Amblystoma* larva 12 mm. in length; oesophageal gland. Apoch. 2 mm., comp. ocular 2.

distal limb, the latter curving cephalad above the ventral posterior margin of the liver, before passing into the midgut.

Four regions may now be distinguished ; a very short anterior region without glands, provided with a ciliated epithelium, a region with flask-shaped glands and ciliated epithelium, a third region with tubular or saccular glands and a mucigenous epithelium, and finally, at the posterior end, a region in which no glands at all are to be discerned. The second and third regions gradually merge into one another, but the posterior non-glandular portion is sharply marked off and forms, in part at least, the future pyloric gland region.

At this stage the two pulmonary diverticula open into a capacious pouch lying below the foregut, into the floor of which it opens. In longitudinal sections the first gland appears immediately behind this sac. Farther back more glands make their appearance, and at the point where the foregut begins to enlarge into the stomach, it is completely encircled by six or eight of these large flask-shaped glands. Farther back again the glands become less and less flask-shaped and take on a tubular or saccular character.

One of these anterior glands is represented in Fig. 4 as seen after staining in haemalum, followed by neutral gentian.



FIG. 4.—*Amblystoma* larva 14 mm. in length; oesophageal gland. Apoch. 2 mm., comp. ocular 2.

The shape of the cells in the body of the gland varies with the degree of distention. There seems to be in these glands an accumulation of the secretion in the lumen distending it, for it is only by the application of a distending force from within that the extreme stretching of the cells, which may be commonly observed, could be produced. In many glands where this distention is great the cells are quite flattened and spread out over a great surface, reminding one strongly of the appearance in the mammalian blastodermic vesicle at the time of its rapid expansion. The explanation there of the flattening of the cells is clearly the stretching caused by the rapid transudation of fluid into

the vesicle, but in these glands it is difficult to explain why the fluid is not discharged into the cavity of the foregut before the pressure gets sufficiently high to cause a stretching of the cells. A possible explanation is the viscosity of the secretion owing to the large number of mucous cells in these glands.

Fig. 4 shows a gland only moderately distended, and here it is seen that the cells at the bottom, where the gland is unsupported by neighboring glands, are drawn out flat, while those at the side still retain their approximately columnar shape.

The two kinds of cells noticed in the earlier larva may still be recognized, the clear mucous cells occupying the top and neck of the flask, the granular cells the sides and base of the flask. The protoplasm of the latter now stains strongly in haematoxylin, and exhibits a faintly striated or finely vacuolated structure. This is due to the presence of prozymogen, which may also be demonstrated by the use of acid alcohol, followed by aqueous haematoxylin after the method of Macallum.¹ The inner end of the cell between the nucleus and the lumen stains but slightly in haemalum, but in sections treated with neutral gentian it is seen to be filled with perfectly round, deeply stained granules of zymogen. The neck of the flask-shaped gland is occupied by long mucous cells of a columnar shape, which also extend into the gland and form the top of the flask. In these cells two zones may be distinguished, an outer protoplasmic, which stains strongly and which contains a quantity of masked iron, and an inner transparent and reticular. The meshes of the latter are filled with a substance which stains faintly in indulin, more readily in Mayer's mucicarmine. In sections stained with neutral gentian many deeply stained granules may be seen in the mucigenous portions of these cells. These are somewhat elongated and not perfectly round, as are the zymogen granules of the other kind of cell. Their significance is not clear; it is possible that they may indicate an imperfect differentiation of the zymogenic and mucigenous functions at this stage of development.

The surface epithelium in this region of the foregut consists of alternate ciliated cells and goblet cells. Tracing the foregut

¹ *Journ. of Phys.* Vol. xxii. 1897.

backward, the glands become gradually tubular or saccular, without any appearance of distention, and the ciliated cells disappear, so that the rest of the glandular portion, as well as all the posterior non-glandular portion of the stomach, is provided with a mucigenous epithelium.

Attention should be called at this stage to the remarkable resemblance between the mucigenous border of the gastric epithelium and the cuticula of the cells in the buccal cavity. Both have a characteristic striated appearance, and one is tempted to think that they cannot be very different chemically.

The cells of the tubular glands do not differ in any respect from those of the flask-shaped glands. The mucous cells are less numerous, and a few glands may be entirely without them. The cells of all the glands, even the very last, contain both zymogen granules and prozymogen.

There are as yet no pyloric glands formed; the epithelium of the posterior portion of the stomach is perfectly smooth and without glandular outgrowths.

Even at this stage there is a remarkable resemblance between the anterior flask-shaped glands and the oesophageal glands of *Proteus* and *Necturus*, and as development proceeds this resemblance becomes more and more striking.

The mouth and pharynx are lined in the aquatic *Amblystoma* larva by a stratified non-ciliated epithelium, with cuticular cells and goblet cells. In a transverse series it may be seen that immediately behind the last gill slit this changes to a ciliated epithelium. One may thus consider the first ciliated cell in a longitudinal section as indicating where the oesophagus begins. Measured from this point the foregut in a larva 16 mm. long is about 3 mm. in length. Of this .49 mm. at the anterior end is non-glandular. Behind this we have a portion .45 mm. long extending from the anterior border of the first gland to a point where the foregut begins to expand to form the stomach. This would doubtless, but for the presence of glands, be regarded as a portion of the oesophagus. Beyond this again, the ciliated epithelium extends into the stomach for a distance of .35 mm. The rest of the stomach is lined by a mucigenous epithelium,

like that of the adult except in its great capacity for division. The posterior portion 1.3 mm. in length is still quite devoid of glands.

Staining with haemalum and neutral gentian shows that at this stage also the cells forming the body of every gland in the foregut contain both abundant zymogen granules and prozymogen, and it is impossible to discern any difference whatever in the cells composing the large anterior flask-shaped glands and the smaller posterior tubular glands respectively, except that in the latter there is no evidence of distention and consequent flattening.

From this stage onward the changes proceed somewhat more slowly and may be summed up briefly.

In a larva 25 mm. in length the foregut measured 5.6 mm. in length. Of this the anterior 1.4 mm. was non-glandular, showing a relatively more rapid growth in length in this portion of the foregut. The ciliated epithelium extended a further distance of .84 mm. into the stomach, the posterior portion of which, 3.36 mm. in length, was lined by the usual mucigenous epithelium.

Fig. 5 is from a larva 32 mm. in length. It is at this stage that the resemblance to the oesophageal glands of *Proteus* and *Necturus* is most marked. The duct of the gland and the portion of the wall nearest to the surface epithelium are composed of elongated cylindrical cells forming a single row. Four of these cells are represented in Fig. 6, *A*, as seen under a high magnification. Each presents an outer granular protoplasmic zone in which the oval nucleus is imbedded, and an inner more extensive zone which is coarsely reticular. They obviously represent the large, clear mucous cells of the ordinary gastric glands, and, as we shall see, are actually transformed into these in the adult. The difference in shape is dependent on external conditions, such as the grouping of the cells, and is not inherent in the cells themselves.

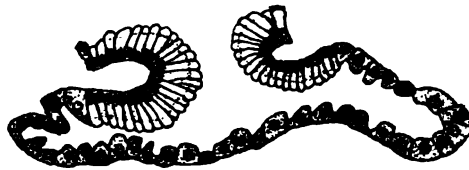


FIG. 5. — Larva of *Amblystoma* 32 mm. in length: oesophageal gland. Ross obj., $\frac{1}{8}$ in., Leitz ocular No. 1.

At the end of the gland they are succeeded suddenly by the zymogenic cells. In these the minute structure is obscured by the large amount of prozymogen present. The cells are less flattened than at the earlier stage of development, probably because the initial distention has been compensated by the rapid division and growth of the cells. A number of these cells is represented in Fig. 6, *B*. They are now somewhat columnar in



FIG. 6. — Oesophageal gland of *Amblystoma* larva. *A*, mucous cells; *B*, zymogenic cells. Zeiss apoch. 2 mm., comp. ocular 8.

shape, with convex ends projecting into the lumen. The nucleus is round or oval and placed in the center of the cell, though in the more columnar cell it is often nearer the lumen than the base of the cell. The free end of the cell may be seen in sections stained with neutral gentian, to be filled with granules of zymogen. Other granules may be seen at the sides of the nucleus, and a few are occasionally found in the base of the cell. The rest of the cell is occupied by a deeply staining protoplasm, which owes its ability to absorb nuclear stains to the large amount of prozymo-

gen present, as may be shown by the employment of Macallum's methods of detecting masked iron. The distribution of the prozymogen determines the appearance of the cell, and three main types are to be recognized; in the first the stain is diffused through the whole of the protoplasm, but more pronounced at the base and sides of the cell, and on close examination a very finely vacuolated structure may be made out; in the second the whole or part of the cell exhibits long deeply staining fibrillae; and in the third type the prozymogen is distributed as small irregularly staining particles throughout the protoplasm.

All the three main types of cells composing the glands and surface epithelium are still capable of division, and numerous mitoses may be seen in all.

Oppel's description of the structure of the oesophageal glands of *Proteus* would apply word for word to these glands

of the larval *Amblystoma*, and in the case of *Necturus* I have satisfied myself, by comparison of the actual objects, that the structures are identical.

The latest larva examined was 65 mm. in length. This animal was apparently about to undergo metamorphosis, as the stratified epithelium of the mouth had been replaced by ciliated. The pyloric glands were fully developed, and the ordinary gastric glands had assumed the appearance they present in the adult. The anterior portion of the stomach, about a millimeter in extent, was still ciliated, but the saccular glands of this region had undergone considerable modification. One of these

is shown in Fig. 7. It will be seen that the base of the gland has grown out into a number of short secondary tubules, formed for the most part of zymogenic cells, and the gland now consists of a large number of such tubules, each similar in structure to an ordinary gastric gland, opening into a large common cavity lined by transparent mucous cells corresponding to the neck cells of an ordinary gland. In short, the saccular gland of the embryo is being transformed into an anterior oxyntic gland of the adult.

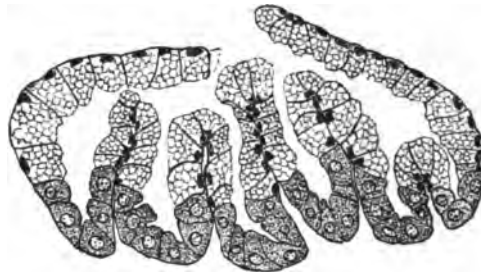


FIG. 7. — Oesophageal gland of 65 mm. larva. Ross obj.
 $\frac{1}{4}$ in., Leitz ocular No. 1.

Two of the most anterior glands in this larva were included in the oesophagus. All were in full physiological activity and were filled with zymogen granules.

I have been unable to secure a specimen of *Amblystoma* undergoing metamorphosis, or one that has just completed it, and am therefore unable to state positively whether all the saccular glands become transformed by subsequent branching into anterior oxyntic glands, or some of them degenerate and disappear. There is in the latest larva that I have examined no evidence of changes of a degenerate nature, and I am therefore inclined to believe that the most anterior glands, as well as the rest, are taken up into the stomach, and that the oesophagus

of the adult is entirely formed by rapid growth from the short non-glandular region of the early larva.

One cannot doubt that the large saccular glands of the larval *Amblystoma* are the homologues of the so-called oesophageal glands of *Proteus* and *Necturus*. The failure of Kingsbury, however, to detect the presence of zymogen granules in the glands of *Necturus* led me to reinvestigate these structures with a view of determining whether or not this was a real point of difference. At first I employed a number of specimens of *Necturus* which had been kept in the laboratory tank for several months without food. In these cases the results were negative, no zymogen granules were present. I afterwards obtained two specimens captured in the vicinity of Toronto, and in a perfect state of nutrition. In these no difficulty was experienced in demonstrating the presence of zymogen granules in the cells of the oesophageal glands. Kingsbury's failure is, in all probability, to be ascribed to the inadequate method he employed to demonstrate the granules. For this purpose he employed treatment of the fresh glands with osmic acid. Now it has been noted by Langley (7) and Grützner (3) that the protoplasm of ferment-secreting cells which contain a great deal of reserve material (prozymogen)



FIG. 8.—Zymogenic cells from oesophageal gland of *Necturus*, showing zymogen granules in free border of cells. Zeiss apoch. 2 mm., ocular 8.

stains strongly in osmic acid. For this reason its use, in cases where the granules are few in number and small, and where there is a great deal of prozymogen present, is of little value. This is precisely the condition in *Necturus*. By the neutral gentian method, however, the granules are stained much more strongly than the prozymogen, and no difficulty is experienced in demonstrating them when present. Fig. 8 shows a number of cells from such a preparation.

It should be added, however, that the oesophageal glands of *Necturus* do not present the evidences of strong functional activity seen in those of the larval *Amblystoma*. The granules are much smaller and may be quite absent from many of the cells of the gland,

even in a well-nourished animal. This is particularly the case in those cells the inner ends of which exhibit signs of degeneration in the shape of the structures described by Kingsbury as mucous globules. It is probable that there is a tendency for these glands in *Necturus* to degenerate rather than remain of physiological importance.

The so-called oesophageal glands of *Proteus* and *Necturus* are really gastric glands the development of which has been arrested. There is also in these animals an arrested development of the foregut, compensation for which has been, in a measure, attained by the conversion of the anterior portion of the stomach into a functional oesophagus. Only a short anterior non-glandular portion actually corresponds to the oesophagus of other Urodela.

Two questions remain to be considered, the relation of these glands to the oesophageal glands of higher vertebrates and to the oesophageal glands of the frog.

The first question is a comparatively simple one. The oesophageal glands of higher vertebrates have no features in common with those of *Batrachia* and are probably of secondary origin. In *Reptilia* oesophageal glands are rare, and where they do occur, as, for example, in *Testudo graeca*, are simple crypts lined by cells similar to those of the surface epithelium, namely, ciliated cells and goblet cells, the latter predominating. In birds and mammals, where the epithelium is usually of the stratified squamous variety, they are more or less complex mucous glands. In no case, as far as I am aware, has investigation revealed in the oesophageal glands of *Sauropsida* or *Mammalia* the occurrence of ferment-secreting cells. It is probable that the oesophageal glands of higher vertebrates have arisen in response to a demand, in a very long and relatively narrow oesophageal tube, for a more efficient lubricating mechanism, and an epithelium that will withstand friction. The first step in this process is the formation of deep crypts lined by ciliated cells and many goblet cells; the second, the disappearance of the ciliated cells from the crypts so as to form a pure mucous gland, and their replacement on the surface by a stratified squamous epithelium.

The second problem is less simple. Because of the very exceptional conditions introduced in the case of the frog by the herbivorous diet of the tadpole, and of the very extensive histolytic changes which take place in the whole intestine during metamorphosis, it becomes difficult to discuss this question from the standpoint of histogenesis. The question is, whether the oesophageal glands of the frog, like those of *Proteus* and *Necturus*, are to be regarded as somewhat modified anterior gastric glands. Let us examine, in the first place, the anatomical characters on which the subdivision of the foregut has been determined in this form. According to Wiedersheim (12) the stomach begins at a point where the foregut experiences an abrupt turn to the left. This is found on examination to correspond to the point where the ciliated epithelium is succeeded by the cylindrical epithelium of the stomach. There is also a slight constriction at this point and a change in the color of the mucous membrane. Of these the only character of importance is the change of epithelium. This is not, in my opinion, a valid criterion for the following reasons: In *Amblystoma* ciliated epithelium is found in the anterior portion of the stomach up to a late stage of development. In the tadpole, according to Gage, the whole foregut is ciliated, and several observers record patches of ciliated cells in the stomach of the adult frog. In several of our American "ganoids," Hopkins (4) and Macallum (9) describe the ciliated epithelium as being continued over a considerable portion of the stomach.

It is true that the differences in the cells of the oesophageal and gastric glands of the frog are very striking; but if we compare the oesophageal glands of the frog with the gastric glands of any *Urodele* or of *Bufo*, these differences are not apparent. The same cellular elements are present, with almost the same arrangement and structure.

The gastric glands of the frog are, in fact, unique among the *Batrachia*, in the small amount of zymogen which they contain. May this not be but another instance in which this animal, as compared with other *Batrachia*, exhibits an unusual degree of specialization, the anterior gastric glands (so-called oesophageal glands) having retained and developed the zymogenic

function at the expense of the oxyntic function, and the posterior the oxyntic at the expense of the zymogenic function, thus foreshadowing in a parallel way the histological differentiation which is seen in the chief and parietal cells of the gastric glands of mammals?

The conditions obtaining in the foregut of *Proteus*, *Necturus*, and the larval *Amblystoma* are of interest apart from their purely histological bearing. For it is obvious that, if the condition in these animals is primitive, the gastric glands of the ancestral types must have occupied a much more extensive portion of the foregut than is the case in existing forms.

Among fishes the subdivision of the foregut into oesophagus and stomach is well marked, not only among the more highly specialized Teleostomes, but also in the sharks and rays. No glands are present in the oesophagus, and the epithelium is different from that of the stomach. In *Amia*, *Lepidosteus*, and *Acipenser*, according to Macallum (9), it is not only extremely difficult to decide on superficial examination where the oesophagus ends and the stomach begins, but on microscopic examination the former is found to have a similar epithelium to the stomach and to contain glands. The nature of these glands is at present in doubt. No doubt the investigation of the structure and histogenesis of the elements of the foregut in these forms, and more particularly in *Polypterus*, will yield highly interesting and instructive results.

LITERATURE.

1. BISCHOFF. "Ueber den Bau der Magenschleimhaut." *Müller's Archiv*, 1838.
2. CARLIER. "The Newt's Stomach during Digestion." *La Cellule*. Tome xvi.
3. GRÜTZNER. "Ueber Bildung und Ausscheidung von Fermenten." *Pflüger's Archiv*. Bd. xx.
4. HOPKINS. "On the Enteron of American Ganoids." *Journal of Morphology*. Vol. xi.
5. KINGSBURY. "The Historical Structure of the Enteron of *Necturus maculatus*." *Proc. Amer. Micr. Soc.* Vol. xvi. Pt. i.
6. KLEIN. "Oesophagus." In Stricker's *Handbuch der Lehre von den Geweben*.
7. LANGLEY. "On the Histology and Physiology of Pepsin-forming Glands." *Phil. Trans. Roy. Soc.* Vol. clxxii.
8. LEYDIG. "Anat. hist. Untersuch. über Fische und Reptilien." Berlin, 1853.
9. MACALLUM. "The Alimentary Canal and Pancreas of *Acipenser*, *Amia*, etc." *Journ. of Anat. and Phys.* Vol. xx.
10. MACALLUM. "On the Distribution of the Assimilated Iron Compounds, etc." *Quart. Journ. Micr. Sci.* Vol. xxxviii. n. s.
11. OPPEL. "Beiträge zur Anat. des *Proteus anguineus*." *Arch. f. Mik. Anat.* Bd. xxxiv.
12. WIEDERSHEIM. *Ecker's Anatomie des Frosches*.

AN EXPERIMENTAL DEMONSTRATION OF THE REGENERATION OF THE PHARYNX OF ALLOLOBOPHORA FROM ENDODERM.

JOHANNA KROEBER.

SEVERAL recent investigators have shown with more or less probability that the lining of the new pharynx which develops during the regeneration of the head in certain earthworms comes from the endoderm, while the pharynx of the embryo is lined by ectoderm.¹ It seemed that by means of experimental methods this relation might be definitely determined. In the following pages I shall describe some experiments on *Allolobophora foetida* that demonstrate, I think, that the lining of the new pharynx is in fact derived from the endoderm.

Hescheler showed, as the result of observations made principally on *Allolobophora terrestris*, that when the five anterior segments are cut off the pharynx is regenerated by a growing forward of the old digestive tract up to the third segment, and that the new buccal cavity occupying the first three segments is formed by an ectodermal invagination. The old pharynx was not completely removed in these operations, since in the normal worm its cavity frequently extends beyond the fifth segment and its thickened muscular dorsal wall always goes back into the sixth, so that Hescheler's results are open to the objection that in his experiments *a part, at least, of the old pharyngeal wall always remained behind* as a possible source for the regeneration of the new pharynx.

Rievel in experimenting on certain Lumbricidae (*Allolobophora foetida*, *Allolobophora terrestris*, *Lumbricus rubellus*) cut off anterior ends consisting of between one-third to two-thirds of the entire body. He arrives at the conclusion that the pharynx is regenerated from the walls of the digestive tract

¹ See Hoffman, *Zeit. f. wiss. Zool.* Bd. lxvi. 1899.

at the point where this was cut, and that no ectodermal invagination whatever occurs, the endodermal diverticulum joining the body wall to form the mouth at the very anterior end of the worm.¹

Haase showed that in *Tubifex*, when four to six anterior segments have been removed, the pharynx grows forward out of the walls of the digestive tract and meets an ectodermal invagination of somewhat varying size. This ectodermal pouch, which forms the buccal cavity, is small in all cases, never extending quite as far back even as the region of the cerebral ganglion.

Von Wagner's observations on *Lumbriculus* show that the point of union of ectoderm and endoderm, originally at the anterior end of the animal, subsequently comes to lie more posteriorly, on account of the forward growth of the "Kopflappen" and accompanying turning in of the ectoderm.

The differences in the accounts cited above show clearly that it is almost impossible to determine with certainty, merely by observation, just how much of the regenerated pharynx ultimately arises from the ectoderm and how much from the endoderm. It is very easy to see where ectoderm and endoderm meet, but the point of fusion is lost soon afterwards, and since the regenerated head continues to increase in size, it is presumably possible that the point of union may come to lie at some distance from its original position. At the time when the pharynx opens to the exterior its walls are not sufficiently developed for one to be able to determine whether the muscles will grow around the endodermal part of the tube; but if in some manner the fusion of the ectoderm with the endoderm could be delayed long enough for the pharyngeal muscles to form around the latter, then the origin of the pharynx might be determined. Hescheler affirms that it is possible to make out the exact limits of the ectoderm by using stains which bring out the cuticle covering this layer. I used the stain which Hescheler mentions as giving the best results, but found that, while my preparations showed in general an agreement

¹ For criticism of Rievel's results, see papers by Morgan (*Roux's Archiv*, Bd. v, 1897) and Hescheler (*Jenaische Zeitschrift*, 1898).

with the figures of Hescheler in regard to the extent of the cuticle on the dorsal wall of the pharynx, they contained also some alternating patches of what seemed to be cuticle and of ciliated areas on the ventral wall and at points in the digestive tract even further back than the regenerated pharynx itself.

For this reason I have attempted to get more certain results by the use of the following experimental methods. Worms, from which the seven anterior segments had been removed, so that no part whatever of the old pharynx was left behind, were allowed to regenerate for a period of between twelve and eighteen days. As a rule the fusion of the ectodermal invagination with the pharynx occurs about fifteen days after the removal of the anterior end of the worm, — although there is considerable individual variation in regard to this point, and also some difference due probably to the temperature, etc. At the end of this time the anterior tip of the new part of the worm was removed in one of two ways: either it was burned off by touching it with a hot needle, or it was cut off with fine scissors. The latter method, though more difficult to carry out successfully, proved to be the better one because the piece cut off could be preserved to show whether the pharynx had joined the ectoderm at the time of the second operation. The worms were once more allowed to regenerate and were finally killed between ten and fifteen days after the second operation. In all cases the worms survived both operations and showed a perfectly normal regeneration, — the only point of difference from worms that had undergone only the first operation being that the new pharynx had time to regenerate before the second ectodermal invagination had fused with its anterior end. The object of the experiment was to determine whether a normal pharynx would develop from the endoderm if the fusion of the ectoderm with it was prevented for a sufficient length of time to allow this development to take place.

It is difficult to determine on the living object whether or not the ectodermal invagination has met the endoderm, and since for my purposes it was best to wait as long as possible before the second operation, it happened in two or three cases,

as sections of the small pieces removed showed, that ectoderm and endoderm had met. In the majority of instances, however, I was fortunate enough to remove the invaginating ectoderm just in time. In cases where this was done with a hot needle there is, of course, nothing to prove that the fusion had not taken place. There is ground for such a belief, however, in the fact that, of a number of worms whose small anterior ends were cut off at the same time after the first operation as when the burning was done, and which were kept under exactly the same conditions, there was not a single one in which the fusion had taken place.

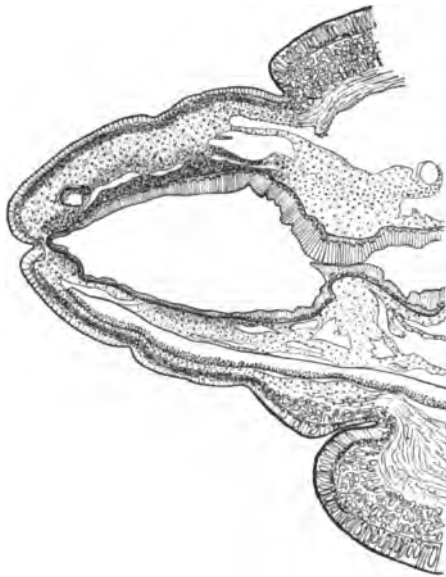


FIG. 1.

The same difficulty presents itself again in determining the time at which the worm is to be killed. I succeeded, however, in getting a number of cases where, though the pharynx and the ectoderm were just on the point of joining, they had not quite done so when the worm was killed. The accompanying figures show two worms in this condition.

Fig. 1 shows a vertical longitudinal section of a worm from which seven segments were removed on January 15. On February 2, that is to say eighteen days later, the tip of the newly regenerated part was cut off. This piece was preserved and sectioned and was found to include the whole of the ectodermal invagination besides the anterior end of the pharyngeal diverticulum which had not yet broken through to the exterior. Fourteen days after this operation, on February 16, the worm was killed.

Fig. 2 represents a vertical longitudinal section of a worm from which the first seven segments were cut off on January

15. The tip of the regenerated part was destroyed with a hot needle on February 2, and the worm was killed on February 17.

Both figures show the diverticulum which has grown out from the walls of the oesophagus about to open to the exterior by fusion with the ectodermal pit; and a comparison with the sections in the same neighborhood shows that these two represent the nearest approach of ectoderm and endoderm to be found in the two specimens. The walls of the pharynx and its musculature, especially on the dorsal side, are well developed. In both worms a nerve cord and a cerebral ganglion have been formed, the latter for the second time. Owing to the slight obliqueness of the section, as shown in Fig. 2, the nerve cord is cut for only a part of its length. The muscles of the body wall have begun to differentiate and there are clear indications of metamerism. All of the worms used in this set of experiments, as well as all those in a later set made to test these results, present a similar condition of things.

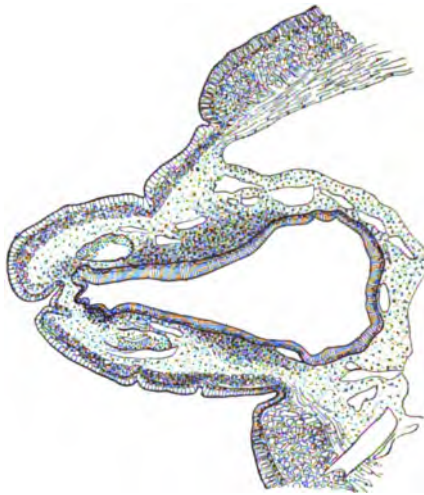


FIG. 2.

From these results we must conclude that the lining of the pharynx is regenerated from the endoderm, while the new ectoderm turns in for a very short distance to meet the pharynx and form the mouth.

The objection may be raised that the possibility of a later pushing in of the ectoderm to form the ultimate lining of the pharynx is in no way removed. But there is no evidence for such an occurrence and, even if it did take place, the fact remains that the musculature of the pharynx develops around an endodermal tube, as my experiments have shown, while in

the embryo the lining of this same region is derived from the ectoderm.

The preceding work was done under the direction of Prof. T. H. Morgan, to whom I wish to express my indebtedness.

BRYN MAWR, May 26, 1900.

FURTHER EXPERIMENTS ON THE REGENERATION OF TISSUE COMPOSED OF PARTS OF TWO SPECIES.

T. H. MORGAN.

THE experiments that I made a year ago were undertaken in order to find out if regenerated tissue, made up of cells derived from two species, showed any mixing of the specific characters of the two species. For this purpose I grafted the tail of a tadpole of one species of frog upon the posterior end of a tadpole of another species. Later the tail was cut off in such a way (as indicated by the line *b-b* in Fig. 5) that the ectoderm left at the exposed edge belonged in part to one species, in part to the other. When the new tail regenerated there was found to be no mixing of the characters of the ectodermal cells along their line of contact in the new part. The results were unsatisfactory

from one point of view, inasmuch as the small piece of ectoderm left after the operation is carried out to the tip of the new tail and increases proportionally less in area than the rest of the new part, so that although it is highly

probable that near the tip of the tail new ectodermal cells are being formed by both kinds of ectoderm, still I did not demonstrate that this is actually the case. Moreover, I found that in the later stages the difference in color between the two kinds of ectoderm was less marked than at first, so that the experiment would have been more convincing had the tail been cut off at an earlier stage. This I have done during the present spring, and the results in regard to the ectoderm

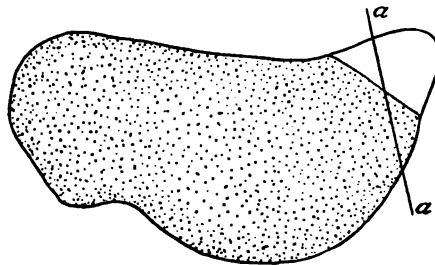


FIG. 1.

confirm in every way those given in my former paper. In the experiments made this spring my main object has been, however, to carry out the experiment in such a way that there would be left at the exposed edge, when the grafted tail was cut off, the internal tissues of two species. In this way I hoped to be able to determine more definitely if, in the newly regenerated part, the tissues mutually influence each other.

The day after the grafting had been performed (*i.e.*, after eighteen to twenty-four hours) the tail was cut off at the region of union of the two components, as shown by the line *a-a* in Fig. 1. In this way there is left at the exposed edge not only the ectoderm of the two species, but the inner tissues also. The regeneration that takes place from the exposed edge will include material derived from both components. Two possibilities presented themselves. First, would the new part be formed of

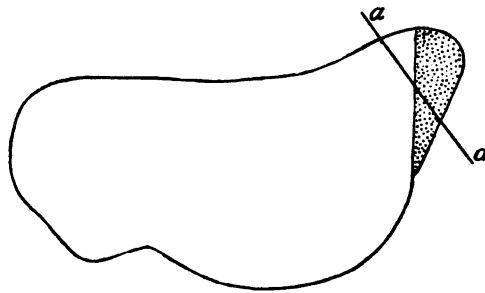


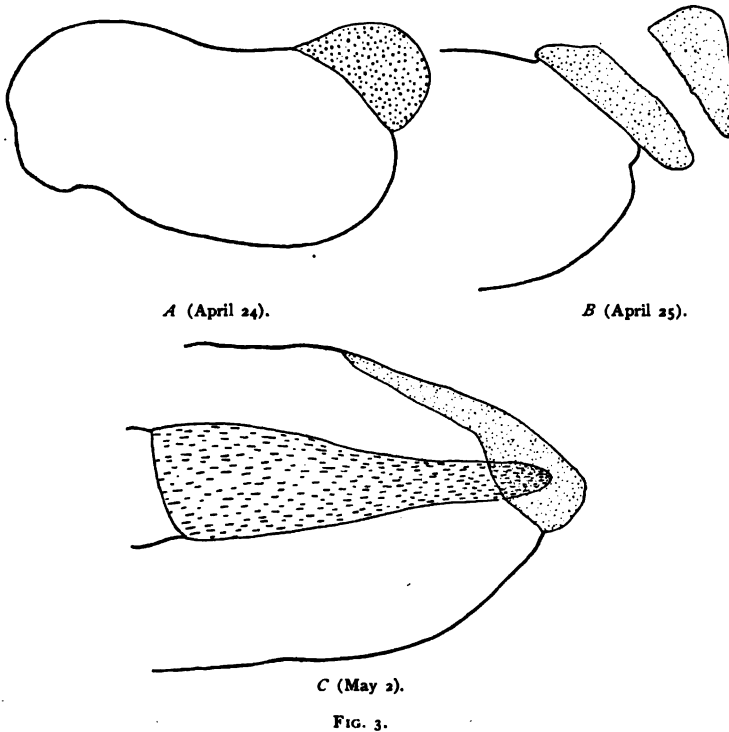
FIG. 2.

cells intermediate in character between the two species as the result of an interaction of the cells on each other; or, second, would the new material preserve the characteristics of the region from which it arises,

or, in other words, one-half of the tail show the characters of one species and the other half of the other species? It is further possible that the new cells might intermingle, and if so the tail might *appear* to be of a hybrid character.

Other experiments of minor interest have also been studied. For instance, in several cases the grafted tail was cut off after twenty-four hours very near its line of union to the major component, as shown in Fig. 3, *A*, *B*. The experiment was made in order to see if the major component might not have some influence on the regenerated part from which it is separated by only a narrow band of tissue of the minor component, but no such influence was observed.

I have found a much safer criterion than before for distinguishing the inner tissues of the two species of tadpoles used in these experiments. In my former experiments I used the differences in color of the pigment cells. I find that this cannot be relied upon under all circumstances. But the muscle tissue of the tail of *Rana palustris* is, especially in the early stages, golden-yellow, while in *Rana sylvatica* the same cells



are slaty-white. The two kinds of cells can be easily distinguished by means of this color difference. Other experiments have shown also that this difference in color is transmitted to the regenerating tissues in the new tail, so that it can be relied upon in the grafting experiments.

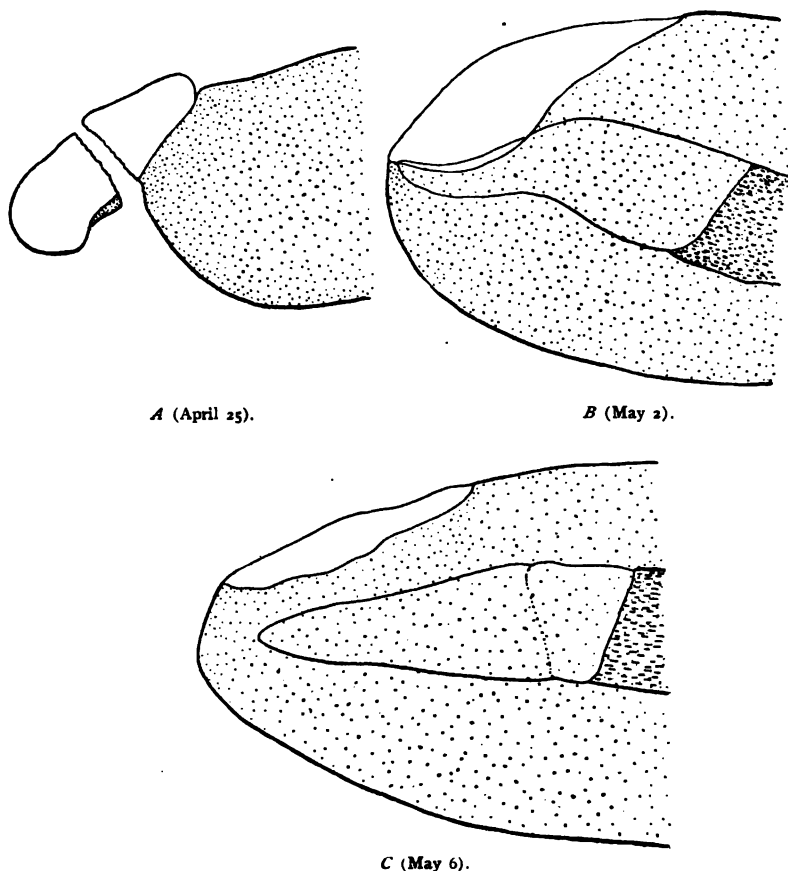
In the first series of experiments the tails were grafted as described in my former paper. After eighteen to twenty-four hours, as a rule, the tail was cut off, as shown by the line *a-a* in Fig. 1. Before grafting it was found more convenient to cut

off the tails obliquely, as shown in the figure—the more anterior end on the dorsal side. Consequently, in order to carry out the second operation of cutting off the tail through the line of union, the cut was made also obliquely, but with the ventral side forward. In a few cases the tails were first cut off with the ventral side further forward (Fig. 2), and the subsequent cutting off was made with the dorsal side forward, as shown by *a-a* in Fig. 2, but the results were practically the same.

It was found easier to graft the tail of *Rana palustris* on the posterior end of *Rana sylvatica* than the reverse. On an average five operations of the former succeeded to one of the latter. The reason for this cannot be given, but it may be due to some difference in the relative sizes of the two components that is more favorable for union in one than in the other way. The result recalls the experiments in cross-fertilization of the eggs in different species, where the crossing can be more easily carried out in one direction than in the other. In this case also the results may be due in some cases to a gross, physical difference, as Pflüger has tried to show for the frog's egg.

In the large majority of cases in which the experiment was carried out as shown in Fig. 1, the core of the new tail seemed to be formed by the minor component, — *i.e.*, if a yellow tail (*R. palustris*) had been grafted upon a black tadpole (*R. sylvatica*) and then after twenty-four hours the tail had been cut off obliquely (Fig. 4, *A*), the central part of the new tail would be composed entirely of the yellow tissue derived from the minor component (Fig. 4, *B*, *C*). The small piece of yellow ectoderm is carried out on the new tail and remains near the tip. It covers a larger area than at first, but it increases not nearly so fast as the rest of the new, yellow tissue of the new tail. The distinctive differences in color can only be seen in the core of the tail, *i.e.*, in the cells that form the muscles. On each side of this axial core a broad fin is present containing inside a gelatinous-like substance with scattered cells, but this fin does not show any difference in color in the two species. It is, therefore, probable that in many cases in which the core of the new tail appears to be composed only of tissue from the minor component that the ventral (or dorsal) connective tissue

of the fin is derived from the major component. The differences in the mesodermal pigment cells are at times very striking, and in all such cases the pigment cells are like those in the tissues from which they immediately arise; but while in many



C (May 6).

FIG. 4.

cases they furnish a safe criterion, in others the difference cannot be made out with certainty. On the other hand, the differences in the muscle tissue of the core can always be seen.

The explanation of the result, *viz.*, that the new tail is in most cases (in forty-seven cases out of sixty) like the minor component, is that in nearly all of these operations too large a piece of the grafted tail has been left. It has contained the

notochord and nerve cord and the tissue immediately around them, and from these the new tail has grown out. I did not discover this until most of my material had been used. After this I cut off the grafted tails nearer the line of union, and although regeneration did not take place so well in several cases, still those that regenerated showed more often both parts contributing to the new tail. The same result followed in a number of cases in the previous operations, and we may now examine how regeneration takes place in such cases.

In thirteen cases (out of sixty) there was found evidence of a dual or compound character to the new tail. In all cases observed there was no evidence to show that the duality was the result of the tissues being mixed in character either by commingling of the cells (each cell retaining its specific characters) or by a hybridizing of the cells (due to mutual influence). The duality consisted in each part, regenerating cells like itself, so that definite regions of the new tail were made up of one or of the other kind of tissue. For instance, the new tail might be made up above of the slaty-colored tissue of *R. sylvatica* and below of the yellow tissue of *R. palustris*. There is no evidence of a shading of one kind of tissue into the other along the line of meeting, but this point would be very difficult to determine positively. There is further no evidence that the two kinds of tissue are any more commingled at the distal end of the tail than at the base.

In regard to the notochord and nerve cord it is extremely unlikely that the cut would ever pass obliquely through the line of union of the one or of the other, as these structures are very small in cross-section. It is, therefore, probable that in nearly every case the new notochord and the new nerve cord are made up of cells belonging entirely to one component. Furthermore, these two structures lie so near together that it is not probable that the cut would pass between them in such a way that the nerve cord at the exposed edge would belong to one component and the notochord to the other component.

The details of the successful experiments are as follows: On April 14 and 15 nine grafts were made, as shown in Figs. 1 and 2. On April 16 these were cut off, as indicated in Fig. 1, *a-a*,

and Fig. 2, *a-a*, but unfortunately the two lots were not kept separately. On April 29 when again examined new tails had begun to regenerate, and two individuals out of the nine showed that the core of the new tail was compound in character. In both the major component was black and the minor yellow. In one of these the new tail was yellow on the dorsal side and black on the ventral, and in the other the new tail was black on the dorsal side and yellow on the ventral.

In another series the experiment was somewhat different. The grafting took place on April 17. Two days later the ectoderm of the minor component had been carried out further on the tail (Fig. 5), so that at the base of the tail the inner tissues of the minor component were covered by the ectoderm of

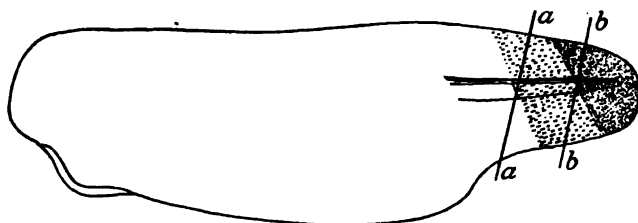


FIG. 5.

the major component. At this time (April 19) the tail was cut off obliquely, as indicated by the line *a-a* in Fig. 4, leaving the inner tissues of both components exposed at the cut surface. On May 19 all three of the tadpoles that had been operated upon showed a compound tail. One of these tadpoles was *R. palustris* and the other two were *R. sylvatica*, with grafted tails of the other species, respectively.

In a third series of five individuals, grafted April 19 and cut off April 20, as in Fig. 1, two showed later a compound tail; and in a third I was in doubt whether or not a few of the yellow cells of the major component entered the new tail.

In three later experiments in which the tail had been cut off, so that a smaller piece of the minor component was left attached, a larger number regenerated compound tails.

In one of these experiments the grafting took place on April 27, and the tail was cut off on the following day. One of the three produced a tail composed of both kinds of tissue.

In another experiment grafted April 28 (2.30 P.M.), as shown in Fig. 2, and cut off April 29 (10 A.M.), two individuals formed abnormal tails and a third a compound tail. The tail of this individual is represented in Fig. 6. On the ventral side of the new tail are found the slate-colored cells of the major component, and on the dorsal side the yellow cells of the grafted piece. (It is not possible to show this difference satisfactorily in a simple uncolored drawing, since the principal difference is one of color.) In addition to this difference one can see in the region at which the grafting took place and where the new tissue arises from the old that each component contributes its half to the new tail. Moreover, in all these cases the tadpoles

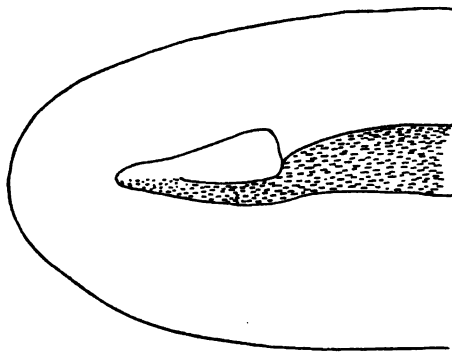


FIG. 6.

had been carefully observed from day to day (and not only at the intervals recorded in the text) and the gradual formation of the compound tail observed.

In another experiment on April 28 the tail was cut off on April 29. One of the tadpoles did not regenerate a new tail,

another (*R. sylvatica*) had a compound tail, and one had a bifid tail, one branch being compound. Finally in another series in which nine grafts were made, one produced a compound tail, another may have contained a small amount of the major component in the new tail, six regenerated entirely like the minor component, and one was abnormal.

In addition to these cases there were three others (in the total of sixty cases) in which there was an overlapping of the two components in the tail, as in Fig. 6. In two of these the core of the new tail came from the minor component, but it is highly probable that a small addition came from the major component also. In the third case the new tail contained at its more distal end elements from both components. Unfortunately this lot was killed accidentally before they regenerated further.

In several cases double tails grew out enclosed in the same common fin, and lay usually in the same plane. In some cases the core of one of the new tails was derived from one of the components and the other from the other component. In several cases one or the other new tail received material from both components. In one of these cases it could be seen with the greatest clearness that the compound tail received material from both sources (Fig. 7).

Cases of this kind are particularly convincing, since they furnish all the data for comparison between the two kinds of regenerating tissue of the two components.

The dorsal tail was yellow and the upper part of the ventral tail was also yellow, and its tissue precisely like that of the dorsal tail. The pigment cells also of the yellow component extended out on to both tails.

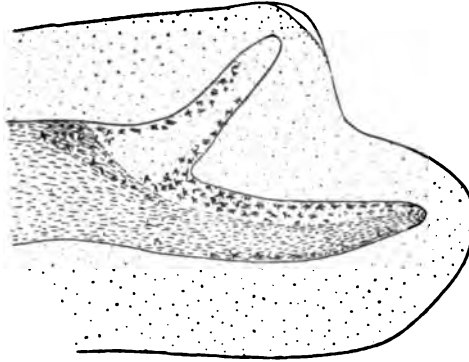


FIG. 7.

These cases of double tails are probably due to imperfect grafting, — the notochord and nerve cord of the two components not being in the same position, so that when the tail is cut off both sets of structures are exposed and a tail develops from each.

Conclusions. — The experiments demonstrate that a single tail may be formed by the regeneration of tissue derived from two species, and that in such cases there is no specific change produced in the one kind of new tissue by the other. Each kind of tissue regenerates its like, and the two kinds combine to form a single morphological organ, — the tail.

May, 1900.

REVIEW OF VON GUAITA'S EXPERIMENTS IN BREEDING MICE.

C. B. DAVENPORT.

IN the two latest volumes of the *Berichte der Naturforschenden Gesellschaft zu Freiburg*, G. von Guaita gives the results of his breeding experiments. He started in 1896 with fifty-five *Japanese walsing* mice and with numerous *white* mice belonging to a race bred by Weismann since 1888, and made crossings through seven generations to 1900. His main data concern the inheritance of color; incidentally, data were got on the diminution of fertility with in-and-in breeding.

Diminution of Fertility.

These results were gained chiefly from Weismann's breedings. The total number of young, the number of litters, and the number of young per litter are given below for each decade of generations.

1st to 10th generation :	1345	young ;	219	litters ;	avg. young per litter,	6.1
11th to 20th "	352	"	62	"	"	5.6
21st to 29th "	124	"	29	"	"	4.2

In von Guaita's breedings :

1st and 2d generations,	.	.	.	.	"	"	"	"	3.5
3rd and 4th "	.	.	.	.	"	"	"	"	3.7
5th and 6th "	.	.	.	.	"	"	"	"	2.9

Thus in the successive generations there is a reduction in fertility of about thirty per cent ; and this is probably due to too close breeding.

Color of Mixtures.

Japanese walzing mice vary in color, but are chiefly piebald—black and white. White mice are without pigment (true albinos) and breed very true.

Crosses of walzing ♀ × white ♂ and white ♀ × walzing ♂ gave twenty-eight young. All were of a gray color and indistinguishable either in respect to color or size from the common house mouse. Also in temperament they were like the house mouse, for they were very wild and lively (unlike either parent) and the walzing action was entirely absent from all the mice of this second generation. Similar results were got by Haacke ('95) after crossing the same races. Haacke says: "When you pair a blue and white spotted walzing mouse with a common white mouse you get either (and usually) uniformly gray mice, which cannot be distinguished from the wild house mouse, or else (more rarely) uniformly black mice." These results, then, lead to the conclusion that when very unlike races of mice are crossed the result is often or usually a *reversion*.

A *third* generation was next produced by von Guaita by mating two of the gray mice or reversions. Four pairs were thus mated and forty-four young were reared—all having both parents gray, and half their grandparents walzing and half white. These forty-four mice are placed in nine color classes, as follows :

				Per cent
"House" or <i>Gray</i> type.	entirely gray,	15	} 25	57
	gray with isolated markings,	7		
	black [essentially house type].	3		
<i>Albino</i> .	pure white, red eyes,	11		25
<i>Walzing</i> type.	white walzers,	3	} 8	18
	gray-white spotted walzers,	1		
	gray walzers,	2		
	black-white spotted walzers,	1		
	black walzers,	1		
		44		100

The most striking phenomenon of this third generation is the sudden occurrence of great variation. In the language of

plant breeders "the *type* is broken." Plant breeders (*e.g.*, Focke and de Vries) have long ago observed that the progeny of hybrids is extraordinarily variable.

Fourth Generation. — Several pairs of the foregoing descendants of the reverted gray mice were mated, and thirty-one young sorted into eight classes were obtained, as follows :

Third Generation ♂	Gray ♀ White- Walzer ♂	Gray ♀ White ♂	White ♀ White ♂	Gray and Spotted ♀ Gray and Spotted ♂	Total	Sum	Per Cent
Uniformly gray . . .	2	12			14		
Gray with markings . .		2		2	4		
Black	2				2	20	65
Albinos	1		4			5	16
Uniformly gray walzers	1	1			2		
Gray walzers with spots		1			1		
Black-white walzers . .	1				1		
Black walzers	2				2	6	19
	9	16	4	2		31	100

As in the third generation, there is here great variation. The results may be generalized as follows :

- (1) All descendants of albinic parents are albinos.
- (2) When both parents are gray and spotted all descendants (2) are gray and spotted.
- (3) Gray ♀ × white ♂ gives 88 per cent gray and 12 per cent walzers ; the white is shut out.
- (4) Gray ♀ × white walzer ♂ gives 44 per cent gray, 44 per cent walzers, and 12 per cent white.

Fifth Generation. — To save room we will henceforth make use of abbreviations for the names of colors, as follows :

G = gray.
 Gw = gray with white markings.
 B = uniformly black.
 Bw = black with white markings.
 A = albino or white.

Ww = white walzers.
 Wg = gray walzers.
 Wgw = gray-white walzers.
 Wb = black walzers.
 Wbw = black and white walzers.

The female parent invariably precedes the male.

Gen.	1	2	3	4	5	6	Total	Sum	Per Cent
III	(G♀ × Ww♂) × (G♀ × Ww♂)	(G♀ × Ww♂) × (G♀ × Ww♂)	(G♀ × Ww♂) × (G♀ × Ww♂)	(G♀ × Ww♂) × (G♀ × Ww♂)	(G♀ × Ww♂) × (G♀ × Ww♂)	(G♀ × Ww♂) × (G♀ × Ww♂)			
IV	Wb♀ × B♂	Bw♀ × B♂	Bw♀ × Wg♂	Gw♀ × Wb♂	Gw♀ × Gw♂	Gw♀ × Gw♂			
G			1				1		
Gw				2	35	16	53		
B		8		1			9		
Bw	2	10		7			19	82	73
A	1	7	1		9			18	16
Ww	1						1		
Wg			1	1			2		
Wgw				2			2		
Wb	1	2					3		
Wbw		5					5	13	11
	5	32	3	13	44	16		113	100

Sixth Generation.

Gen.	Colors				Total	Sum	Per Cent
II	All gray or reversions						
III	A, 2; Gw, 4; G, 2	$\left\{ \begin{array}{l} A, 1; Gw, 2; \\ G, 3; Ww, 2 \end{array} \right\}$	Gw, 2; G, 3; Ww, 3	G, 4; Ww, 4			
IV	Gw, 4	$\left\{ \begin{array}{l} Gw, 1; Bw, 1; \\ Wg, 1; Wb, 1 \end{array} \right\}$	$\left\{ \begin{array}{l} Gw, 1; Bw, 1; \\ B, 1; Wb, 1 \end{array} \right\}$	Bw, 1; B, 2; W, 1			
V	Gw $\times$ Gw	Wgw $\times$ G	B $\times$ A	Wb $\times$ B	A $\times$ Ww		
G	1	1				2	
Gw	20	1				21	
B			7	1		8	
Bw			16	1		17	48
A	10		18		4		38
Ww			2			2	
Wb		1				1	3
	31	3	43	2	4		83
							100

Seventh Generation. — The colors of only eight members of this generation were determined — too few to be significant.

General Results.

In the successive generations the percentage of walzing individuals undergoes a steady decline from eighteen per cent and nineteen per cent in the third and fourth generations to eleven per cent in the fifth and four per cent in the sixth generation. Is this decline due to the elimination of an unstable condition or to the circumstance that too little of the walzing blood has been employed in the later crosses to keep up the original proportion? The question whether the normal law of inheritance is followed here may, indeed, be asked of all the colors. The normal law of inheritance, as defined by Galton, is that one-half the heritage of any generation is derived from the parents, one-fourth from the grandparents, one-eighth from the great-grandparents, and so on, according to the formula :

$$\text{Inheritance} = \frac{1}{2} k_1 + \frac{1}{4} k_2 + \frac{1}{8} k_3 + \frac{1}{16} k_4 + \text{etc.}$$

To apply the normal law of inheritance it is convenient to assume it and to compare the theoretical heritage with the empirical. If the two agree, the validity of the law is established in this case; conversely, if the two do not agree, the law does not hold. This method of testing the law is the same as that employed by Galton ('98) in the case of Bassett hounds. It may be illustrated by the calculation of the theoretical number of albinos in the sixth generation. Let us take the first column. If one of the two parents were an albino, we should expect at least $\frac{1}{2} \times 50\%$ of the progeny to be such. If both parents were A, $\frac{3}{4} \times 50\%$ of progeny at least should be A. If, in addition, all of the grandparents (Gen. IV) were A, we should expect at least $\frac{3}{4} \times 50\% + \frac{1}{4} \times 25\%$ of the VI Gen. to be A. In general, if n_v, n_{iv}, n_{iii} , etc., represent the number of times an albino appears as ancestor in the different generations, then the proportion of albinos in the sixth generation should be :

$$\%A_{vi} = \frac{n_v}{2} \times 50\% + \frac{n_{iv}}{4} \times 25\% + \frac{n_{iii}}{8} \times 12.5\% + \frac{n_{ii}}{16} \times 6.25\% \\ + \frac{n_i}{32} \times 3.125\% + \frac{1}{2} \times 3.125\%.$$

The last term is got by observing that the ancestors of half of the first generation were exclusively albinos for many generations, while the ancestors of the other half were exclusively walzers. The value of A is similarly calculated for each column, and the theoretical number of individuals for each column is found. Their sum should be equal to the observed number, or, when reduced to percentages, to the observed percentage of total. The closeness of theory to observation is sometimes striking. Thus if we compare column by column the observed and theoretical frequencies of walzers in the fifth generation we get :

COLUMN	1	2	3	4	5	6	TOTAL	PER CENT
Observed	2	7	1	3	0	0	13	11
Calculated	2.19	6.00	1.13	4.88	0	0	14.19	12.5

In the following table are given for each generation the observed and (in parenthesis) the corresponding calculated percentages for each color :

GENERATION	I	II	III	IV	V	VI
Black alone (B) . . .	{ 0	0 (0)	7 (0)	7 (0)	25 (18)	30 (40)
Gray less black (G) {	0	100 (0)	50 (50)	58 (48)	48 (60)	28 (28)
Total gray and black {	0	100 (0)	57 (50)	65 (48)	73 (78)	58 (69)
Albinos	{ 50	0 (50)	25 (25)	16 (32)	16 (9)	38 (18)
Walzers	{ 50	0 (50)	18 (25)	19 (20)	11 (13)	4 (13)

Several remarkable things come out of this table. In the first place the most marked departure from Galton's Law of Ancestral Inheritance is seen in the second generation, where the gray, non-walzing reversioners suddenly made their appearance. We know as yet little concerning the laws of the phenomenon called "reversion"; but whether it be considered a remote atavism or only an apparent "inheritance," it seems equally to form an exception to Galton's Law.

Secondly, the case of the walzers does indeed look like an exception to Galton's Law. It looks as though the walzing condition were an unstable condition being rapidly eliminated. In so far the result opposes the usual expectation of sport prepotency.

Thirdly, the albinos, likewise sports, apparently are prepotent, since there is twice the proportion there should be in the sixth generation. The numbers are so large that one can hardly object that these figures are not altogether significant.

Fourthly, the grays run close to theory, excepting always generation II. They are nearest to the original type of *Mus musculus* and seem to inherit in the most nearly normal fashion.

In conclusion, then, we may say that the data afforded by these breeding experiments indicate, so far as they go, that Galton's Law of Inheritance holds only with form units which are not very divergent from the type, and that among sports we may have some that show a great stability and prepotency, while we may occasionally have others which are physiologically so unfit that they are unstable and have less than normal potency.

LITERATURE CITED.

- FOCKE, W. O. ('81). Die Pflanzen-Mischlinge. Berlin, Borntraeger, 1881. 569 pp.
- GALTON, F. ('97). "The Average Contribution of Each Several Ancestor to the Total Heritage of the Offspring." *Proc. Roy. Soc. London*. Vol. lxi, pp. 401-413.
- GUAITA, G. VON ('98). "Versuche mit Kreuzungen von verschiedenen Rassen der Hausmaus." *Ber. Naturf. Ges. zu Freiburg*. Bd. x, 3 Heft, pp. 317-332. April, 1898.
- GUAITA, G. VON ('00). "Zweite Mittheilung über Versuche mit Kreuzungen von verschiedenen Hausmausrassen." *Ber. Naturf. Ges. zu Freiburg*. Bd. xi, 2 Heft, pp. 131-138. August, 1900.
- HAACKE, W. ('95). "Ueber Wesen, Ursachen und Vererbung von Albinismus und Scheckung und über deren Bedeutung für vererbungstheoretische und entwicklungsmechanische Fragen." *Biol. Centralbl.* Bd. xv, pp. 44-78. 1895.

VARIATION IN THE TEETH OF NEREIS.

MARY HEFFERAN.

THE purpose of this quantitative study in variation is to determine the modal condition of a character in a species of *Nereis* commonly found on the west coast of the Atlantic. The material was very generously placed at my disposal by Professor Charles B. Davenport, who collected it during the summer of 1899 at Cold Spring Harbor, Long Island. The animals were found in the sand of the beach and were taken at random, excepting that small ones were rejected.

These annelids went by the familiar name of *Nereis virens*, but upon comparing them with Ehlers's (68, p. 559) descriptions and drawings of that species, I found that although they agreed in most characters, an important difference occurred in the length of the first or postoccipital segment; that of *N. virens* being twice as long as the second segment, while that of the Cold Spring Harbor form is about equal to or even slightly less than the second in length. In this character, as also in that of certain parapodal bristles, the "Sichelanhänge," which are rather short and broad instead of slender and long as in *N. virens*, the Cold Spring Harbor species agreed well with Ehlers's description of *N. limbata*, the distribution of which also includes the west Atlantic coast. From these two points, and from the fact that Cold Spring Harbor is slightly south of the southern limit described for *N. virens*, and within the range of *N. limbata*, it seems probable that we are here dealing with the latter of Ehlers's two species. It may be possible that the two species overlap in this region at the southern limit of *N. virens*, and that my collection contained both. However, nothing in the numerical results of my investigation seemed to suggest two distinct or even transitional forms.

1. *Method.*

The specific character selected for investigation was the number of teeth on the jaw. This number is commonly stated by authors in descriptions of species.

The jaws are two in number, from 1 to 3 mm. in length, brown, horny, curved, and serrated along the inner or falcate margin. They are at the extremity of a large exsertile proboscis which is usually retracted in alcoholic specimens, so that in order to free the jaws it is necessary to cut down the median line of the head, ventrally, through to the inside of the muscular proboscis. By turning the head backwards the jaws can be made to extrude, and the teeth counted by means of a hand lens. In the specimens killed later in the season by a slowly killing fluid, the jaws remained extruded.

In counting, some difficulty was experienced in fixing a limit in either direction, at the curved, distal end in those cases in which very fine teeth ran to the tip, and particularly at the proximal extremity where the outlines of the teeth were indefinite. In order to count these it was necessary to pull out the jaws gently with a forceps, and to free the bases from connective tissue, carefully, without entirely separating the jaws from the head. Since the line of division between the free, prominent teeth and the undeveloped ones, buried in a chitinous band, was always distinct, the method was adopted of counting them separately, and of including in each set all that showed well-formed outlines when held up against a strong light and viewed through a lens from the dorsal side. Those connected by the chitinous band were called indefinite teeth, the rest the definite teeth. Totals were found by adding.

2. *Results.*

I. TABLE OF DISTRIBUTION OF FREQUENCIES.

Classes	1	2	3	4	5	6	7	8	9	10	11	12	13	14
L. Def. . .		5	30	88	128	102	41	6						
L. Indef. .		12	30	93	146	93	21	4	1					
L. Total .						3	12	28	95	116	100	39	7	
R. Def. . .	1	4	37	94	126	86	41	11						
R. Indef. .	2	7	30	80	149	97	28	7						
R. Total .					1	2	8	27	102	114	89	46	10	1

From the table of frequencies it will be seen that the number of definite teeth varies from 1 to 8, the indefinite from 1 to 9, the total numbers from 5 to 14.

The following constants were obtained.

II. TABLE OF CONSTANTS.

	LEFT DEF.	LEFT INDEF.	LEFT TOTAL.	RIGHT DEF.	RIGHT INDEF.	RIGHT TOTAL.
n	400	400	400	400	400	400
M	5	5	10	5	5	10
A	5.098 ± 0.040	4.905 ± 0.039	10.00 ± 0.044	5.043 ± 0.043	5.013 ± 0.040	10.055 ± 0.045
σ	1.193 ± 0.028	1.179 ± 0.0279	1.306 ± 0.031	1.267 ± 0.030	1.191 ± 0.028	1.339 ± 0.032
F	$+0.7025$	-0.66148	$+0.0599$	$+0.4339$	-0.8617	-0.5314
Type	I	IV	I	I	IV	IV
Skewness	-0.0369	-0.0439	-0.1341	$+0.0153$	-0.0868	-0.0509

Comparison of these numerical results suggests the following conclusions :

The mode, 5, is the same for the definite and indefinite on both sides, and for the total on each side it is 10. The averages also show little difference between the right and left jaws. From the variability, however, as indicated by the standard deviation, it appears that the number of teeth of the right jaw is slightly more variable than that of the left, σ being 0.074 greater on the right side for the definite teeth, 0.011 (which is less than the probable error) for the indefinite teeth, and 0.033 greater for the right total. The highest degree of variation is shown by the total number of teeth on the right side, and the least variation is shown by the number of indefinite teeth.

The form of the distribution curve of definite teeth on both jaws falls under Pearson's Type I, with the peculiar result of a slight negative skewness for the left side, and a positive skewness, although a very slight one, $+0.0153$, for the right. This indicates a tendency to the production of fewer definite teeth than normal on the left side, and a faint tendency towards a greater number on the right side. The distribution curve of indefinite teeth on both sides is of Type IV, with a negative skewness, -0.043 , on the left side, and twice this, -0.086 , on the right ; *i.e.*, there is on the right side a greater

tendency than on the left towards the production of few indefinite teeth. On the right side, then, it is clear that the probability of a large number of definite teeth is associated with that of a small number of indefinite teeth. The same thing is shown for the left side, for although definite and indefinite teeth both show negative skewness, the negativeness is much greater in the indefinite than in the definite teeth. Therefore, relatively, the skewness of the indefinite and the definite teeth may be said to be, here also, of opposite sense. This agrees with results shown in the correlation table, to be noted later.

A peculiar result is obtained in regard to the distribution curve of the total number of teeth. The left total falls into a curve of Type I, while the right total is of Type IV. The negative skewness of the latter is -0.050 , while that of the former is about two and one-half times as much. The table of frequencies shows that the right total includes two classes more, one at each end of the series, than the left total. There is one individual in each of these two classes. It seemed probable, by inspection of the calculation, that the critical function, F , which was negative 0.5314 , might be made positive by dropping these two extreme individuals, thus giving a curve of Type I. I found this to be the case, and obtained for F the value $+0.210$; but I found further that Type I might be obtained by dropping only the individual of Class 5, making $F +0.0389$. The skewness in this case was very slight, only -0.00706 .

In order to determine which was the closer fit of the observed curve to the theoretical curve in the two types, I calculated the theoretical curves from the observed data with the following result.

TYPE IV.¹

$n = 400$	$d = 0.06822$	$M = 10.055$
$s = 25.66$	$m = 13.830$	$y_0 = 96.34$
$a = 6.5798$	$\sigma = 1.3386$	zero ordinate = $9.1114 (M-md)$
$v = 3.6796$	$\phi = 8^\circ 9' 7''$	$\tan \theta = \pi/a$
	$y = y_0 (\cos. \theta)^{2m} e^{-v\theta}$	

¹ For the methods of calculating the results given in the following tables, see Davenport, '99, pp. 20, 23, 24.

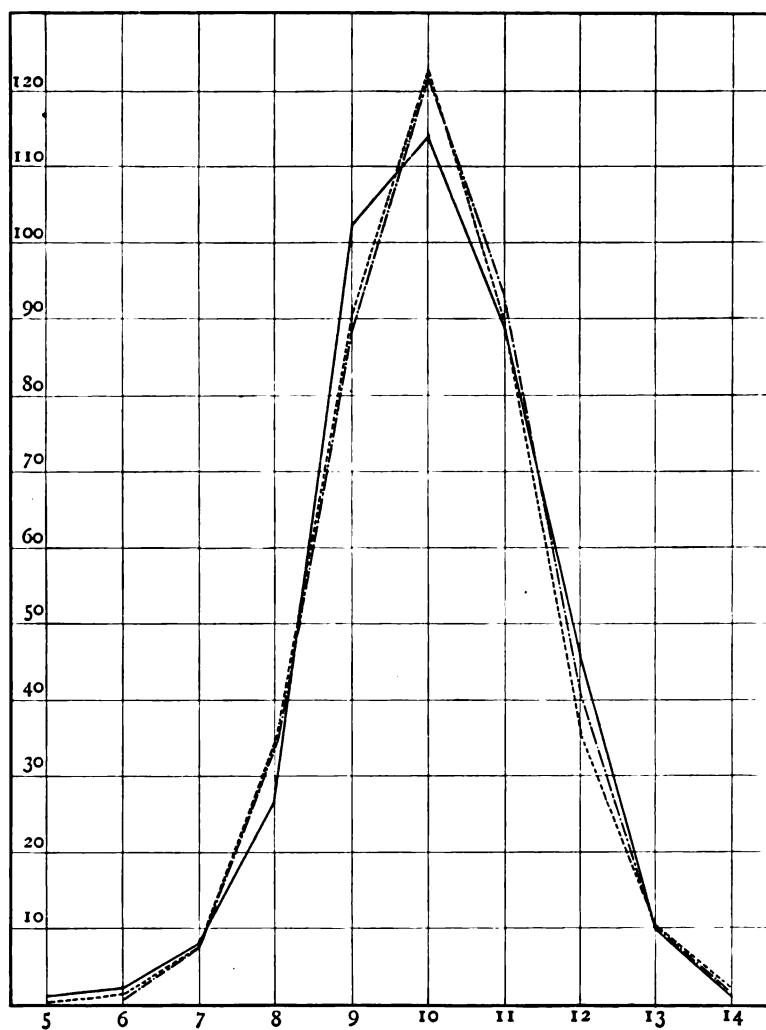


PLATE I.

- Polygon of observed frequency.
- Polygon of theoretical frequency, Type IV.
- Polygon of theoretical frequency, Type I.

Δ , Calculated by Duncker's method, = 3.68%.

$V-M$	f	y	δ_1	δ_2
- 4.1114	1	0.1	+ 0.87	
- 3.1114	2	1.2	+ 0.8	
- 2.1114	8	7.9	+ 0.1	
- 1.1114	27	35.3	- 8.3	- 0.09
- 0.1114	102	90.5	+ 1.5	- 4.82
+ 0.8886	114	122.9	- 8.9	- 5.01
+ 1.8886	89	89.9	- 0.98	
+ 2.8886	46	35.5	+ 7.43	- 0.85
+ 3.8886	10	10.8	- 0.89	- 0.79
+ 4.8886	1	2.3	- 1.36	
	400	399.7	41.05	11.56

TYPE I.

$n = 399$	$s = 305.56$	$b = 46.165$
$a_1 = 21.663$	$m_1 = 142.426$	$\sigma = 1.316$
$a_2 = 24.502$	$m_2 = 161.134$	$d = 0.00929$
$y_0 = 120.68$ (calculated from approximate formula). $M - d = 10.066$		

$$y = y_0 \left(1 + \frac{x}{a_1}\right)^{m_1} \left(1 - \frac{x}{a_2}\right)^{m_2}$$

Δ , Calculated by Duncker's method, = 3.58%.

$V-M$	f	y	δ_1	δ_2
- 4.066	2	0.9	+ 1.07	
- 3.066	8	7.8	+ 0.2	
- 2.066	27	35.2	- 8.24	- 0.19
- 1.066	102	87.2	+ 14.84	- 5.29
- 0.066	114	121.5	- 7.46	- 4.96
+ 0.934	89	93.1	- 4.08	
+ 1.934	46	41.1	+ 5.06	- 2.25
+ 2.934	10	10.3	- 0.3	- 0.29
+ 3.934	1	1.4	- 0.42	
	399	398.5	41.60	12.98

Since it is considered a sufficient agreement between observation and calculation when $\Delta < \frac{100}{\sqrt{n}}\%$, which in this case is 5%, it is evident that these values show a moderate degree of closeness of fit of the two curves. The difference between the two values of Δ , 0.10, is so small that we may conclude that it is practically immaterial under which type this curve should fall.

The fact that by dropping one individual from a series a curve may be thrown from Type IV to Type I and may be made to fit equally well in either case, raises a serious question as to the biological importance of the distinction between Pearson's Type I and Type IV. Pearson ('95) himself says: "It seems very possible that discreteness rather than continuity is characteristic of the ultimate elements of variation; in other words, if we replaced the curve by a discrete series of points, we should find a limited range. It is the analytical transition from this series to a closely fitting curve which replaces the limited by an unlimited range. Exactly the same transition occurs when we pass from symmetrical point binomial to normal curve. Thus while Type I marks an absolutely limited range, Type IV does not necessarily mean that the range is actually unlimited."

It appears from the results obtained in the two calculations given above that even less value can be placed upon any distinction between Type I and Type IV than is suggested by Pearson. The difference of one individual actually causes, here, the transition from one type to the other, the individual being at the extreme of the series.

3. *Correlation.*

In the table on the following page every possible combination of teeth for the two sides is given, together with the actual number of specimens for each combination of definite, indefinite, and total teeth.

From this series of combinations the following results were obtained for the coefficient of correlation. The calculations were made by Pearson's method and checked by the briefer method of Duncker.

Correlation between Right and Left Definite Teeth,					$r = +0.688 \pm 0.0136$
"	"	"	"	" Indefinite "	$r = +0.725 \pm 0.0121$
"	"	"	"	" Total "	$r = +0.820 \pm 0.0081$
"	"	Right Definite and Left Indefinite,			$r = -0.424 \pm 0.0231$

Bearing in mind that an index of 1 signifies a perfect correlation, and that 0 indicates an entire lack of it, we see that the

III. CORRELATION TABLE.

Teeth.		Specimens with definite teeth.	Specimens with indefinite teeth.	Teeth.		Specimens with definite teeth.	Specimens with indefinite teeth.	Teeth.		Specimens with total teeth.	Teeth.		Specimens with total teeth.
R.	L.			R.	L.			R.	L.		R.	L.	
1	2		2	5	5	67	79	5	6	1	11	9	1
1	4	1		5	6	26	20	6	6	2	11	10	20
2	2	2	5	5	7	6	2	7	7	7	11	11	60
2	3	1	2	5	8		1	7	8	1	11	12	7
2	4	1		6	3	2		8	7	4	11	13	1
3	2	2	4	6	4	8	5	8	8	13	12	9	2
3	3	14	12	6	5	17	32	8	9	8	12	10	5
3	4	14	11	6	6	49	54	8	10	2	12	11	14
3	5	6	2	6	7	9	7	9	8	11	12	12	23
3	6		1	7	4	1		9	9	61	12	13	2
3	7	1		7	5	4	5	9	10	25	13	12	6
4	2	1	1	7	6	13	15	9	11	5	13	13	4
4	3	7	9	7	7	21	8	10	7	1	14	12	1
4	4	41	39	7	8	2		10	8	3			
4	5	36	27	7	9		1	10	9	23			
4	6	9	4	8	6	3		10	10	64			
5	3	6	8	8	7	5	3	10	11	21			
5	4	22	38	8	8	3	3	10	12	2			
						400	400						
													400

degree of correlation between the right and left sides is, on the whole, rather high. It seemed at first a somewhat unexpected result that the correlation in the variability of what I have called the indefinite teeth should be higher than in that of the definite teeth. If the correlation had been perfect it would have meant that those causes which produced a deviation from the mean in the left sets acted in the same degree on the right sets also. Is it possible then that different causes may have acted or that the same cause may have been effective in different degrees in producing the variability in the definite and the indefinite teeth?

This question drew my attention more closely to a fact noticed only incidentally in counting the teeth, *i.e.*, in case of animals having dark, heavy jaws, evidently older animals, the definite teeth were fewer in number than in case of small, young individuals. In the older jaws the teeth began farther from the tip, leaving a smooth point, while the younger, more

delicate jaws were often finely denticulated to the extremity. The serratures of the older jaws appeared to be worn off by use. In order to determine whether or no the correlation actually existed between the size or age of an animal and the number of definite teeth, I made the comparison for one hundred individuals, taking as an indication of size, and hence roughly of age, the head length in millimeters. This was measured from the anterior edge of the first ring to the extremities of the two apical feelers. The result was a negative correlation, although a rather small one, -0.128 . It is probable, then, that age does come in as a factor in the production of a small number of teeth, and that this decrease is due to wear. It is possible also that the wear may be heavier upon one jaw than upon the other, thus accounting for the comparatively slightly lower degree of correlation between the definite teeth than between the indefinite teeth. For wearing does not act at all directly on the indefinite teeth, since they do not emerge from the chitinous covering, and are often sunk in the tissue of the proboscis. It would be interesting to know in what manner the jaws are carried and work upon each other during the life of the animal, for a certain habit of crossing them might also account for the peculiar differences in skewness of the curves of the right and left teeth noted in the discussion of constants. The smallness of the negative index of correlation between the age of an animal and the number of definite teeth shows that age does not play a very important part in causing variation.

An attempt to correlate the number of definite and the number of indefinite teeth on the right jaw resulted in a negative index of correlation, -0.424 . This fact indicates an inverse relation between the definite and indefinite teeth on the same jaw; that is, a jaw with a small number of definite teeth will probably have a comparatively large number of indefinite teeth, and inversely. It may be that indefinite teeth continue to be laid down at the base of the jaw during the life of the animal, in which case the number would tend to be greater with age, while, as we have seen, the number of definite teeth is smaller.

4. *Relation of Individual and Specific Variation.*

Out of fifty different species of *Nereis* which I found described by various authors, the number of teeth was stated for forty-seven. The numbers ranged from 0 in two cases, in which the edentulous condition of the jaws was an important specific character, to 20, given by Audouin and Milne-Edwards ('29) for *N. fucata*. The number of teeth of *N. fucata* is given by Ehlers ('68), however, as 7, by Johnston ('65) as 5 to 10, and in the Challenger ('85) reports as 14 to 16. It would be difficult here, as in the case of a few others, to decide which observer came nearest to the modal condition of the species. It is also impossible to tell whether they counted the total number of teeth including those covered by a chitinous band, or whether they referred only to the prominent definite teeth. Ehlers makes the distinction only in *N. virens*, where he gives the definite teeth as 5 to 6, total as 10. St. Joseph ('88, '98) notes in description of *N. diversicolor* that of 8 teeth 2 are indefinite, and in *N. floridana* that of 9 teeth the lower 4 are buried in a clear translucent covering. For these two species, respectively, Ehlers has given 8 and 9 teeth, evidently counting both definite and indefinite.

After attempting various methods of striking averages of the statements made by different authors I finally decided to use Ehlers's numbers alone as most reliable, adding a few of those given by St. Joseph in which there was less doubt that the total number of teeth had been counted. Seriation of twenty-two species gave the following results, total teeth.

Classes . . .	5	6	7	8	9	10	11	12
Frequencies	1	1	5	2	6	3	2	2

CONSTANTS.

$A = 8.727$	$F = +1.354$
$\sigma = 1.838$	curve = Type I
$\beta_1 = 0.000090$	$s = 5.862$
$\beta_2 = 2.323$	skewness = +0.00966

Any conclusions which can be drawn from these results are necessarily of doubtful value. It will be seen that the mean

for the number of teeth in twenty-two species is lower than the mean of total teeth in the one species which I have described. The skewness of the curve instead of being negative is positive, although it is exceeding small. Had it been negative, as I had thought it might be, it would have indicated that in the species of the genus, as well as in the individuals of the species, there had been a movement in the direction of a smaller number of teeth, either from an excessive production of individuals and species having few teeth, or from selective annihilation of those having many teeth. The opposed positive skewness is so small that it may mean little in regard to the species, and particularly since the numbers are small and the method of counting so doubtful no stress can be laid upon it.

5. *Abnormalities.*

Differences between the two jaws of the same animal in the definite, indefinite, or total number of teeth were of common occurrence. The accompanying drawings are intended to show some irregularities of this kind. In Fig. 1 the right jaw has four large definite teeth and five below which do not emerge from the surrounding chitinous layer; the left jaw has only slight crenulations corresponding to five definite teeth, although it has the same number of indefinite teeth as the right side. Fig. 2 shows on the right jaw three definite teeth, the edge above and distal to them having three very slight elevations; the opposite jaw ends in a long point with a perfectly smooth edge and has only two large definite teeth below. There is the same number of indefinite teeth on both sides. Figs. 3 and 4 show variations of the same kind, the numbers of both definite and indefinite teeth being different for the two jaws.

So far the drawings have been made from old animals in which the jaws are hard, strong, and very dark in color. It is probable from the appearance of the jaws that the difference in the number of definite teeth is due largely to the wearing, on one side or the other, of the distal teeth. Fig. 5 shows a common irregularity of equal totals with slight differences in

the combinations of definite and indefinite teeth, the left jaw having 8 to 4, the right 6 to 6. Figs. 5 and 6 are from small, young animals, and the jaws are seen to be more slender with numerous fine teeth, 13 on the right and 12 on the left jaw.

The specimen drawn in Fig. 7 was interesting in regard to the indefinite teeth. The left jaw presented the usual appear-

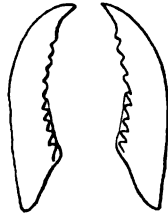


FIG. 1.

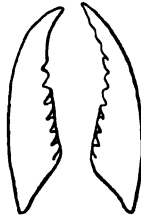


FIG. 2.

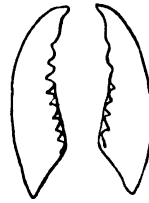


FIG. 3.

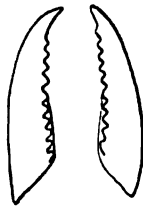


FIG. 4.



FIG. 5.



FIG. 6.

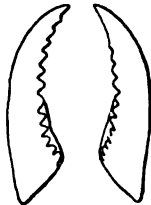


FIG. 7.

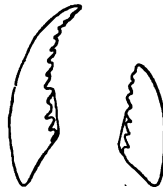


FIG. 8.

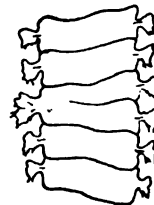


FIG. 9.

PLATE II.

FIGS. 1-8. — Variation and abnormalities in teeth on opposite jaws.

FIG. 9. — Abnormal segment.

ance of six indefinite teeth placed fairly regularly and six definite teeth. On the right jaw the point was worn smooth, leaving only four definite teeth, while below three normal indefinite teeth was a series of five small ones placed very close together instead of three as on the opposite jaw. This may indicate a tendency towards regulation by the production

of an excess of teeth at the base of a jaw on which some of the extreme teeth had been lost, but I found no other indication of such regulation. Another individual presented a partial right jaw, Fig. 8, which was a stump of about half the length of the left jaw. The normal jaw was dark brown, almost black, while the stump was light straw color characteristic of a young jaw or of the very base or imbedded part of an old one. The color indicated new growth or regeneration, in which case one would expect to find a production of small indefinite teeth crowded at the base, as in the specimen of Fig. 7, if that could be interpreted as a regenerative process. On the contrary, the stump had exactly the same number of teeth similarly disposed as the part of the opposite jaw which corresponded to it. It may have been, then, only the rounded stump of a broken jaw, although this explanation does not account for the peculiar color.

Abnormalities in other parts of the animals were looked for only incidentally. I found no cases of double pairs of caudal cirri, but all of the worms were not examined for this not unusual abnormality, since the posterior parts of many of them were not preserved.

Fig. 9 shows a case of an abnormal segment. The fifteenth segment extended only a little more than halfway across towards the left side of the body, leaving one broad segment on the left side in place of two, and a partially double parapod. The axis of the body was bent at that point, compensation being made gradually by the greater width on the left side of the segments immediately preceding and following.

6. Summary.

The results of this study may now be summed up as follows :

(1) The typical condition for the total number of teeth of *N. limbata* of Cold Spring Harbor, 1899, is a curve of either Type I or Type IV, with a slight skewness in a negative direction from the mode, 10.

(2) In case of the calculation of the right total teeth, a transition from a curve of Type IV to an equally serviceable

one of Type I could be made by discarding one extreme individual out of four hundred.

(3) The number of teeth on the right jaw appears to be slightly more variable than that on the left.

(4) The degree of correlation between the two jaws is, on the whole, rather high, 0.820. Correlation is closer between the indefinite than between the definite teeth. An inverse relation exists between the number of definite and the number of indefinite teeth on the same jaw, and also one between the number of definite teeth and the age of an animal.

(5) The class range of teeth as given by the different species of the genus *Nereis* has a close agreement with the class range of *N. limbata*. Hence this one species offers the material for the modal condition of all species of the genus.

(6) The results of observations of many specimens showing irregularities in teeth point to the conclusion that a process of wearing away of the definite teeth takes place in mature animals, and therefore that age comes in to help produce small number of teeth, but is not a large factor in causing variation.

Only one author, St. Joseph, makes note of a difference between young and old specimens, the young having the greater number of teeth. Thus the statements made in regard to the number in many species in which only one animal or at most very few specimens were seen and described by their discoverers, are of little value as criterions of the specific condition.

In conclusion, I wish to express my thanks to Professor Charles B. Davenport, who not only generously furnished the material for this investigation, but by his oversight and advice greatly facilitated the progress of the work.

BIBLIOGRAPHY.

1. AUDOUIN ET MILNE-EDWARDS ('29). "Classification des Annélides, et Description de celles qui habitent les côtes de la France." *Ann. des Sci. Nat.* 1^e série, tome xxix, pp. 195-221.
2. DAVENPORT, C. B. ('99). *Statistical Methods, with Special Reference to Biological Variation.* New York, 1899. 148 pp.
3. EHLERS, ERNST ('68). *Die Borstenwürmer. Zweite Abtheil.* Leipzig, 1868.
4. JOHNSTON, GEORGE ('65). *A Catalogue of the British Non-Parasitical Worms in the Collection of the British Museum.* London.
5. M'INTOSH, WILLIAM C. ('85). "Report on the Annelida Polychaeta Collected by H. M. S. Challenger." *Challenger Reports.* Vol. xii, pp. 210-230.
6. PEARSON, K. ('95). "Skew Variation on Homogeneous Material." *Phil. Trans. Roy. Soc. London.* Vol. 186 A, p. 389.
7. ST. JOSEPH, M. LE BARON ('88, '98). "Annélides Polychètes des côtes de Dinard." *Ann. des Sci. Nat., Zool. et Paleon.* 7^e série, tome v, pp. 266-269. 8^e série, tome v, pp. 288-304.

BIOLOGICAL BULLETIN.

THE CENTROSOME IN THE MATURATION AND FERTILIZATION OF *BULLA SOLITARIA*.

MARTIN SMALLWOOD.

THE material upon which the following observations were made was collected at Woods Holl during the seasons of 1898-1900. The greater part of the work was done at the Marine Biological Laboratory under the direction of Dr. E. G. Conklin, and I take this opportunity of thanking him for his many valuable suggestions. I also wish to acknowledge my indebtedness to Dr. C. O. Whitman and Dr. C. W. Hargitt.

A fuller account of my observations both upon the subject of this paper and the cell lineage of *Bulla*, with a discussion of the pertinent literature, will be published later.

The sketches illustrating the mitotic changes in maturation were drawn from sections stained with Heidenhain's iron-alum followed by an aqueous solution of Bordeaux. In order to differentiate the sperm, it was necessary to use Conklin's mixture of haematoxylin and picric acid. The sperm, therefore, has been drawn from corresponding stages and inserted into these figures.

In the interpretation and nomenclature of the centrosome and sphere I have followed in the main Van Beneden. The term "centrosome" will be applied to the body which occurs at the pole of the spindle, etc., when that body has become differentiated into a central corpuscle, surrounded by a clear area, the medullary zone bounded by a definite wall. The body occurring at the center of the aster is the central corpuscle.

The central corpuscle becomes the centrosome. Sections of the ovotestis before copulation show the unfertilized egg lying free in the follicles of the hermaphroditic gland. The large germinal vesicle lies in the center of the egg; it contains a large vacuolated nucleolus, also basichromatin and oxychromatin granules. The deutoplasmic spheres are equally distributed in the cytoplasm and conceal its structure. I have not been able to discover any evidence of a central corpuscle or centrosome in the egg before mitosis begins. By the time the eggs are laid the first polar spindle is in the end of the prophase. In order to secure the earlier stages, a large number of animals were collected and killed as soon as they began to lay. The first polar spindle begins to form as the animals begin to lay. Sections of the ovotestis taken from animals killed while they were laying revealed the fact that every mature egg had already passed through the early prophase of the first spindle; even those eggs in the most distant follicles, where it is prob-

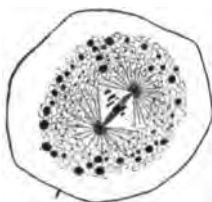


FIG. 1. — Taken from the ovotestis. Shows the central corpuscle surrounded by the cortical zone of the sphere. The central spindle is well formed and the chromosomes are forming into the first equatorial plate. Walls of the germinal vesicle still present, sperm entering at vegetal pole.

able that the sperms from the receptaculum seminalis had not penetrated; I have found the sperms in the anterior part of the hermaphroditic duct, but not extending back to any considerable distance. The earliest stage thus far discovered had two well-formed central corpuscles and a definite central spindle connecting each, which passed through the germinal vesicle, the walls of which are plainly visible. The ring-shaped chromosomes have begun to form from the meshwork of linin and chromatin. The reduction of the chromosomes has not been worked out in detail. These ring-shaped chromosomes gradually take a deeper stain

and come to lie in the equatorial plate of the first polar spindle (Fig. 1). In a cross-section of the equatorial plate I was able to count sixteen distinct chromosomes. There is a distinct cortical zone surrounding the central corpuscle. The astral rays pass through this clear area and extend to the central corpuscle. At this stage I have been able to

trace them nearly to the egg membrane. From now on, there does not appear to be any appreciable change in the mitotic figure, until after the egg has been laid, when it begins to migrate to the periphery of the egg, where it assumes a radial position. There is no difference in the character of the two poles of the spindle. During this movement of the spindle the chromosomes pass into the metaphase, and the centrosome becomes differentiated into a central corpuscle and a medullary zone which is limited by the walls of the old central corpuscle. This is the first time that we have a structure to which we can apply the term "centrosome" in the sense that I purpose to use the term. The cortical zone has enlarged and become much fainter. The chromosomes do not divide at once; the activity is centered in the centrosomes. While the chromosomes are still in the equatorial plate, the central corpuscle in each centrosome divides, having the dumbbell form. The centrosome rapidly increases in size, the periphery is limited by a definite line, which gradually becomes thinner. The medullary zone does not take a plasma stain, as it did in the previous stage (Fig. 9). The centrosome now begins to assume an elliptical form and at the same time to rotate. This rotation continues until the elongated centrosome, which encloses the second polar spindle, lies radially and in the same position that the first polar spindle did. The central corpuscles, connected by a central spindle, are so influenced by this elongation of the walls of the centrosome that they come to lie near the ends — at the foci of the ellipse. As the outer pole of the second polar spindle nears the periphery of the egg, the rays extend to the chromosomes, and they are pulled into the spindle to form the equatorial plate of the second polar spindle. It will be seen that in the main my results corroborate those of MacFarland,¹ Lillie,² and Conklin.³

The changes which take place in the centrosome during this stage are very interesting. The centrosome is so large and comes out with such perfect clearness that I have been able to

¹ "Celluläre Studien an Mollusken-Eiern," *Zoöl. Jahrb.* 1897.

² "Centrosome and Sphere in the Egg of *Unio*," *Zoöl. Bull.* Vol. i, No. 6. 1898.

³ *Science*, March, 1898.

follow the details very carefully. The spindle is developed when the central corpuscles separate. At this time a very faint line can be seen connecting the new corpuscles; as the distance between them increases, the line becomes more distinct, until a central spindle can be clearly distinguished. In the mean time the line limiting the centrosome has become broken into pieces, which gradually become smaller and smaller until they cannot be distinguished from the granules of the cytoplasm. While these changes have been taking place, this broken line has served to mark the outer limit of the medullary zone. The old medullary zone has disappeared, and between

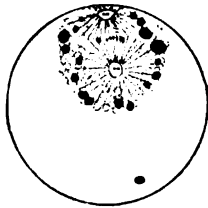


FIG. 2. — Metaphase of first maturation spindle. Two central corpuscles at each pole. Cortical zone limited by a dotted outline. The medullary zone does not take a plasma stain at this stage. Sperm head solid and elliptical.

the central corpuscle and the broken wall of the centrosome we have a new medullary zone, which is the cortical zone of the second polar spindle. The process is as follows: as the walls of the centrosome begin to break down, an area next to the central corpuscles and at each end of the centrosome appears; this begins to take a plasma stain (Fig. 3). The area gradually surrounds the central corpuscle and all of the space at the end of the spindle between the central corpuscle and the wall of the centrosome. In the mean time the central corpuscle has increased in size and is to become the cen-

trosome of the second polar spindle; it ultimately becomes differentiated into a central corpuscle and a medullary zone.

Fig. 2 shows two central corpuscles in the centrosome at each pole of the spindle. Linville¹ shows a similar but not identical stage. The centrosome of the outer pole of the spindle does not enlarge more than is shown in the figure; as it reaches the surface of the egg, it breaks down and the central corpuscles form the division centers in the first polar body.

There is no telaphase in the first maturation spindle. The nearest approach to such a stage is shown in Fig. 3, where the

¹ "Maturation and Fertilization in Pulmonate Gasteropods," *Bull. Mus. Comp. Zool., Harvard*. Vol. xxxv, No. 8.

chromosomes have become partly hollow vesicles. A few of the interzonal fibers show at this stage, but they are very faint.

In the metamorphoses of the centrosome its attachment to the astral rays is plainly evident; the old rays can sometimes be seen in a stage younger than the one shown in Fig. 11, when the new rays have already begun to form and are attached to the central corpuscle. I believe that the rays of the first polar spindle disappear and that the rays of the second spindle rise *de novo*.

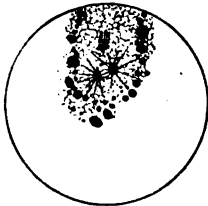


FIG. 3.—Prophase second maturation spindle. Wall of centrosome broken in pieces. Central corpuscles connected by a spindle. New cortical zone forming. Chromosomes are partly hollow. A few interzonal fibers are present.

The metakinesis of the second polar spindle takes place very rapidly. The chromosomes elongate, divide transversely, and as they move toward the poles, they assume a roundish form, and change into vesicular bodies which fuse to form the female pronucleus. During the time when they are fusing, the rays can be traced directly into the areas immediately surrounding them. In the stage of anaphase as represented in Fig. 5, the centrosome is evident, although it does not stain as deeply as in Fig. 4. Immediately after this stage the centrosome disappears and the cortical zone enlarges and completely surrounds the female pronucleus; later both male and female pronuclei come to lie in this clear area. A single centrosome passes off with the second polar body, which is much smaller than the one given off in the first polar body (Fig. 4).

The eggs of this species are not especially favorable for a study of the problem of fertilization. During all of the earlier stages, the sperm head lies completely surrounded by deutoplasmic spheres. I have not been able to make out any continuous clear area about the sperm head during its progress through the egg. In one instance there was a definite clear area about the sperm nucleus after it had nearly approached the female pronucleus, otherwise it was unattended

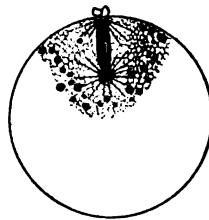


FIG. 4.—Anaphase second maturation spindle. Centrosome at each pole with a single central corpuscle. The medullary zone takes a plasma stain.

by anything that corresponds to the "Hellerhof" of MacFarland and others. The sperm enters at the vegetal pole, though not in any definite place. The tail is lost before the sperm enters the egg membrane (Fig. 1). The head is a solid body having a distinct angle in the middle. If there is a middle-piece, it is practically indistinguishable. The only indication that I have found of such a body is that on one end of the sperm head sometimes one finds a narrow band that stains a little denser than the rest of the head.

The sperm head becomes top-shaped as it begins to migrate toward the animal pole with the point leading. The head

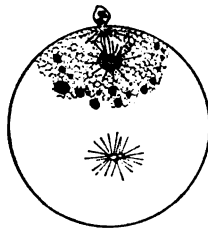


FIG. 5. — Late anaphase of second maturation spindle. Centrosome still present. Cortical zone enlarging. The sperm is composed of three vesicles and accompanied by astral rays.

becomes elliptical (Fig. 3) and stains very black. It remains in this solid form for some time, while the first polar spindle is passing from the metaphase until the anaphase of the second polar spindle. During the anaphase of the second maturation, the solid sperm head becomes vesicular, and for a very short time is accompanied by astral rays. I have not been able to discover a central corpuscle in connection with the aster, nor have I ever seen an amphiasster. At this same time secondary asters usually appear in the egg, which are smaller than the

sperm aster. As the chromosomes of the second polar spindle begin to assume the vesicular form, the sperm aster disappears, and the sperm, consisting of one or more vesicles, rapidly approaches the inner pole of the second polar spindle. When the sperm consists of more than one vesicle, these fuse into one when the aster disappears. While the vesicular sperm is shifting its position it does not increase in size to any noticeable extent, but as soon as it comes near the female pronucleus, which now consists of but three or four vesicles, it rapidly increases in size until it is about twice as large as it was when migrating toward the animal pole. From the time that the sperm head enters the egg until it comes to lie in contact with the female pronucleus (Fig. 6), it is not attended, so far as I have observed, by any body which might be taken for a central corpuscle or a centrosome.

The structure of the two pronuclei when they have come together (Fig. 6) is the same. The male pronucleus is usually regular in outline and slightly smaller. The irregularities

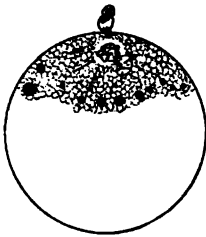


FIG. 6. — The female pronucleus is irregular in outline, surrounded by the cortical zone of the inner pole of the second maturation spindle. A few astral rays are present. Zwischenkörper of second polar body present.

in the outline of the female pronucleus often persist until the central corpuscles of the first cleavage appear. The chromatin stains very slightly and is connected by delicate linin threads. The changes through which the chromatin passes before the equatorial plate is formed may be described under three stages. First, the chromatin rapidly increases in staining power, forming a dense reticulum, often containing stellate masses of solid chromatin. Second, the chromatin begins to assume a definite form. The most conspicuous shape is the stage where the masses of chromatin have begun to break up

into rings but are still attached to one another. The chromosomes have not yet become hollow. They stain uniformly. Third, the chromosomes have broken apart from each other, and have become hollow, round bodies. At first there is a delicate meshwork connecting them (Fig. 7), but this is soon lost and each pronucleus is filled with ring-shaped chromosomes. While the chromatin is undergoing the last two changes, the central corpuscles (the so-called cleavage centrosomes) of the first cleavage spindle make their first appearance. I have found them in a much earlier stage than the one figured, but in each case there was no connection between them; but these corpuscles with their rays have a definite relation with the pronuclei, that is to say, each pronucleus has an aster and central corpuscle with a faint cortical zone. My observations upon the cleavage centrosomes would tend toward the position, first, that they arise *de novo*; and, second, that one arises in connection with each pronucleus.

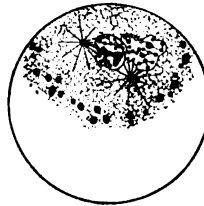


FIG. 7. — Origin of cleavage centrosomes. One in connection with each pronucleus. The central corpuscles are surrounded by a faint cortical zone.

Metamorphosis of the Centrosome in Maturation.

The definiteness and clearness with which the several changes in the centrosome appear in *Bulla* make these changes the most important of the various stages in maturation and fertilization. In describing the changes of the centrosome, under various stages, I have no theoretical points in consideration. While the stages figured are clearly differentiated, still there are intermediate stages which graduate imperceptibly into one another.

In the earliest prophase that I found the central corpuscle was a large solid mass (Fig. 8). Surrounding the central corpuscle there was a conspicuous area, the cortical zone, which was sharply differentiated from the cytoplasm. The rays are not lost in the cortical zone, as MacFarland has shown for *Dialula*, but extend to the central corpuscles, as Lillie has shown for *Unio*, and Linville for *Limnaea*. However, I do not find a row of microsomes, as in *Unio*, limiting the sphere, nor is the boundary formed by the fusing of the astral rays, as in *Limnaea*.

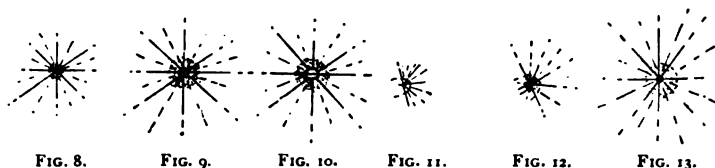
Second stage (Fig. 9). The central corpuscle has become clearly differentiated into a centrosome. It reacts to stain in a very different manner from what it did in a previous stage. There is now a medullary zone which takes on a plasma stain and is limited by a distinct line. The small dark body in the center is the new central corpuscle. The cortical zone has increased in size and is less easily distinguished from the surrounding cytoplasm. The line marking the periphery of the centrosome is the limiting wall of the enlarged central corpuscle of the previous stage.

Third stage (Fig. 10). The centrosome has increased in size. The line at the periphery is definite and whole. The central corpuscle of the previous stage has divided into two central corpuscles, which are connected from the first by faint lines. The medullary zone does not take a plasma stain. The cortical zone has become very faint and soon disappears as a distinguishable area in the cytoplasm.

Fourth stage (Figs. 3 and 11). The periphery of the centrosome loses its continuity, and openings occur in the wall; while

these changes in the wall are taking place, the centrosome becomes much enlarged and assumes an elliptical shape. Immediately after these breaks appear, there is a small part of the medullary area which takes a plasma stain. This area is somewhat triangular in shape and occurs at the end of the centrosome between the central corpuscle and the broken periphery of the centrosome. New astral rays are formed which extend to the central corpuscle. The old astral rays can be seen disappearing at this stage in the cytoplasm.

Fifth stage (Fig. 12). The central corpuscle of the second polar spindle has enlarged and is still undifferentiated. The pieces of the periphery of the old centrosome have become



FIGS. 8-13. — The changes through which the central corpuscle and centrosome pass from the prophase of the first maturation spindle to the metaphase of the second maturation spindle. In each case the solid dark body is the central corpuscle. The granular area surrounding the central corpuscle is the cortical zone. Figs. 9, 10, and 13 show a complete centrosome, having a central corpuscle and medullary zone.

smaller. There is now a distinct cortical zone around the central corpuscle, which has been derived from the medullary zone of the centrosome of the first polar spindle. This stage is identical with the first one described, except that the rim of the old centrosome is still present and the central corpuscle is only about one-half as large.

The sixth stage (Fig. 13) shows the rim of the old centrosome still present, but in smaller pieces which do not stain as deeply as in the previous stage. The cortical zone has enlarged and become fainter. The centrosome is composed of a medullary zone and a central corpuscle.

Summary.

The central corpuscle of the first polar spindle becomes the centrosome of the second polar spindle with a medullary zone and a central corpuscle. The medullary zone of the centro-

some of the first polar spindle gives rise to the cortical zone of the second polar spindle. The central corpuscle of the centrosome of the first polar spindle gives rise to the centrosome of the second polar spindle. Thus we may say that the centrosome of the first polar spindle in *Bulla solitaria* not only gives rise to the centrosome and mitotic figure of the second polar spindle, but to the cortical zone or outer sphere substance surrounding each centrosome.

ALLEGHENY COLLEGE,
October, 1900.

CONTRIBUTIONS ON THE MORPHOLOGY OF THE ACTINOZOA.

J. PLAYFAIR McMURRICH.

VI. *HALCURIAS PILATUS AND ENDOCOELACTIS.*

IN 1892 Carlgren showed that certain Edwardsiae, whose tentacles were more numerous than the mesenteries, had these tentacles arranged on the hexactinian plan, their arrangement in this presumably primitive group of the Actiniaria seeming to foreshadow what is characteristic of the phylogenetically later group. In other multitentaculate Edwardsiae he found what seemed to be an octamerous arrangement combined to a certain extent with hexamerism, but later studies ('99) convinced him that the octamerism did not occur, and that in all cases the hexamerous arrangement obtained.

In the mean time an important discovery had been made by Fautot ('95) in studying *Edwardsia beautempsii* and *E. adenensis*, the former of which possesses fourteen to sixteen tentacles, while for the latter the number is stated to be fifteen to sixteen. Sections through the column showed the eight mesenteries, which have long been supposed to be the only mesenteries developed in the Edwardsiae; but in the uppermost portions a number of additional very short and narrow mesenteries were found which in *E. beautempsii* were placed in such a way as to make with the perfect mesenteries an arrangement recalling what occurs in *Gonactinia prolifera*. Thus there were eight pairs of mesenteries present in the upper part of the column, two of which, the directives, were formed of two perfect mesenteries, four of one perfect and one imperfect mesentery, and one of two imperfect mesenteries. In *E. adenensis* the additional short mesenteries were arranged in pairs in each interval between adjacent perfect mesenteries, except in the endocoel of the directives, so that in this form the arrangement differed somewhat from that typical for the hexactinians.

These observations show reason for believing that in the Edwardsiae there is an intimate relation between the number of tentacles and that of the mesenteries, and that when there are more than eight tentacles there is a strong probability that a number of short mesenteries are also present in the upper part of the column. It is a general rule in the Actininae that the number of tentacles in the fully developed condition is double that of the pairs of mesenteries or, in other words, that there is a tentacle corresponding to each endocoel and each exocoel, the number of tentacles being equal to the total number of mesenteries. Exceptions, due to a lack of development of the full complement of tentacles, are of common occurrence, in many cases probably owing to the specimens examined not having reached their full development, though even in some adults, apparently, the number of tentacles never reaches that of the mesenteries, as is the case, for instance, in *Peachia hastata*, which, with twenty mesenteries, never has more than twelve tentacles.

The rule may be better expressed by saying that *the number of the tentacles never exceeds that of the mesenteries*, and when an apparent exception to this occurs the presumption is that closer examination will reveal the existence of small mesenteries limited to the upper part of the column and in sufficient numbers to fulfill the requirements of the rule.

Acting on this supposition, I have made a further study of the upper portion of the column of *Halcurias pilatus*, a form which I have already described as possessing twenty mesenteries and a number of tentacles considerably in excess of that of the mesenteries, having been estimated in one specimen ('93) to be about seventy, and in another ('98) to be about sixty. Sections showed, as I had expected them to do, the presence of a number of short and narrow mesenteries in the upper part of the column, the number of these *plus* the twenty perfect mesenteries being equal to the total number of the tentacles, which proved to be sixty-eight.

The sections also revealed, however, a peculiarity which I had not expected, and which, as may be seen from Fig. 1, consisted in the short, narrow mesenteries being developed in the

endocoelic spaces bounded by the perfect mesenteries. The sections did not, unfortunately, cut the column perfectly transversely, but the arrangement which obtained may be perceived from the representation of the half of one section shown in Fig. 1, and from the diagram (Fig. 2) which represents a reconstruction from a perfect series of sections. On each side of the median line of Fig. 1 is one of a pair of directives, that to the right being cut at the level of the oral stoma, as is also another mesentery in the right half of the figure. On each side of the



FIG. 1. — Transverse section through the upper part of the column of *Halcurias pilatus*.
T = tentacle. D = directive mesenteries.

directives is a perfect mesentery with its muscle pennon on the same side as that of the adjacent directive, and on the left side this mesentery is succeeded by one which evidently forms with it a typical pair, though it may be noticed that the endocoel enclosed by this pair is broader than the adjacent exocoels. On the right side, where, owing to the obliquity of the sections, the column is cut higher up, the bases of some of the tentacles (T) being cut, the mesenteries of the first lateral pair are widely separated, and between them three imperfect pairs occur, which evidently represent two cycles. The succeeding

exocoel is much narrower than either of the adjacent endocoels and contains no imperfect mesenteries, while in the next endocoel two pairs of mesenteries are seen on the right side and one on the left. By following through the series of sections it is readily seen that the arrangement found in the first lateral endocoel of the right side is repeated in all the others, except in the cases of the endocoels enclosed by the directives, and

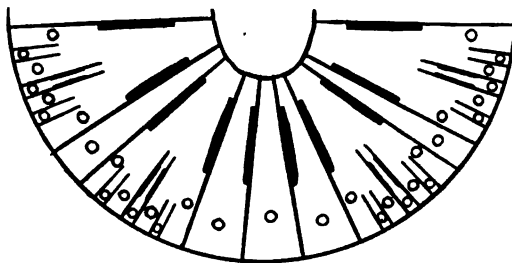


FIG. 2. — Diagram showing the arrangement of the mesenteries and tentacles in *Halcurias pilatus*.

the condition represented diagrammatically in Fig. 2 is that which obtains.

Owing to the relative widths of the endocoels and exocoels, and the presence of imperfect mesenteries in the former, the first impression one receives is that of a form with a large number of directive mesenteries. That such an interpretation of the conditions is erroneous is clearly shown, however, by reference to the mesenteries on either side of the true directives. It is interesting to note that the development of the imperfect mesenteries, which are plainly arranged in two cycles, follows the hexactinian rule, the smaller pairs being developed in the intervals between the larger pairs and the adjacent perfect mesenteries. It may be added that my sections show the existence of a marginal stoma in each perfect mesentery in addition to the oral stoma already mentioned.

From the description given above, it will be perceived that the arrangement of the mesenteries in *Halcurias pilatus* is identical with that described by Carlgren ('97) for a form from the Chinese seas which he refers to the genus *Endocoelactis* and to a new family, the Endocoelactidae. The similarity to *Halcurias* is by no means confined, however, to the arrangement of the mesenteries, and there can be no question but that the two forms must be referred to the same genus, to which, notwithstanding the greater appropriateness of Carlgren's name, the prior term, *Halcurias*, must be applied. The

specific identity of Carlgren's form with *H. pilatus* seems improbable; for, apart from the difference in the localities for which the two have been obtained, the tentacles of the Chinese form are longer apparently than those of *H. pilatus*, and to judge from Carlgren's figures, the longitudinal musculature of the tentacles is weaker and its mesogloal processes coarser. It seems preferable at present to regard them as distinct, and since Carlgren, in his brief notice, has bestowed no specific name on his *Endocoelactis*, I would suggest that it be named *Halcurias Carlgreni*, as a slight recognition of the admirable work which that author has accomplished on the morphology of the Actiniaria.

An examination of the arrangement of the tentacles of *H. pilatus* with reference to the mesenteries was made in the series of transverse sections and also by an examination of the disk, and the results obtained were essentially the same as Carlgren's. I was not able, however, to distinguish any difference in the position of the tentacles over the endocoels bounded by the imperfect mesenteries, though on theoretical grounds it is probable that some difference does exist, and, furthermore, the study of sections seemed to indicate that the tentacles over the directive endocoels were situated a little nearer the mouth than were the others represented as being in the same cycle in Fig. 2; an examination of the disk failed, however, to confirm this appearance.

As regards the systematic position of *Halcurias*, a few remarks are in order. I at first ('93) assigned it to the family Halcampidae, but later ('98) deemed it advisable to separate it from that family and refer it to Hertwig's Antheomorphidae. Carlgren in the mean time had established for his *Endocoelactis* the family Endocoelactidae. There are apparently three courses open for the disposal of the genus. It may be referred to a family already existent, the definition of the family being changed, if necessary, to accommodate it; or it may be taken as the type of a distinct family, as Carlgren has done; or, finally, it may be separated altogether from the Hexactiniae and regarded as the type of a separate tribe.

It seems to me that this last procedure is quite unnecessary,

and would probably be entirely out of harmony with the phylogenetic relationships of the genus. We have learned within recent years how extensively nearly allied forms may differ, and how great are the modifications which the hexactinian type may undergo. The entire facies of *Halcurias* is that of an hexactinian, and it may furthermore be pointed out that instances of the occasional endocoelous development of mesenteries have been already recorded by G. Y. and A. F. Dixon ('89) in *Bunodes thallia* and by Haddon ('98) in *Actinioides dixoniana* and *A. papuensis*.

If, then, the third possibility be excluded, *Halcurias* must either be assigned to an existent family, the endocoelous development of mesenteries being regarded as of minor importance, or this feature may be considered of sufficient importance to warrant the establishment of a separate family. I have already indicated my belief that the peculiar mode of development of the secondary and tertiary mesenteries is of minor importance and see no more reason for separating *Halcurias* as the type of a new family than I do for separating an octamerous sagartian, or one with a multiplicity of mouths and many siphonoglyphs, from the rest of the members of that family.

It remains then to consider what the forms may be with which *Halcurias* may be associated. As Carlgren has remarked, and as I have indicated by the position to which I have referred it in previous papers, *Halcurias* occupies a position near the base of the hexactinian stem. The small number of perfect mesenteries, the occurrence of reproductive organs on all of them, the absence of a distinct sphincter muscle, the simplicity of the margin, are features which, when combined in one individual, indicate for it a somewhat low position. Are there other forms which present a similar combination of peculiarities associated with the development of an adherent base?

Two forms suggest themselves in this connection, namely, *Gonactinia prolifera* and *Protanthea simplex*; but both of these present peculiarities which render their association with *Halcurias* inadvisable. They both have but eight perfect mesenteries, the remaining ones, eight in *Gonactinia* and about

eighty-eight in *Protanthea*,¹ being imperfect, and the ciliated lobes are lacking in their mesenterial filaments. On account of these peculiarities it seems to me that these two forms must be grouped together in a family, Gonactiniidae, as Carlgren ('93) has proposed, and *Halcurias* cannot be placed with them. The family Gonactiniidae must, I believe, be placed among the Hexactiniae, as indeed must all the forms which I have included in the past in the order Protactiniae, as well as those which Carlgren has referred to the Protantheae. The discovery of hexactinian mesenteries in certain Edwardsiae, already referred to, necessitates either the abolition of both this order and that of the Protactiniae, or else an extension of the latter to include both the Edwardsiae and many of the Halcampidae, and it seems to me that the former step is the most practical and the most in accord with a correct phylogenetic scheme. Not that I mean by this that the stages of development shown by the members of the group do not represent phylogenetic stages in the evolution of the Hexactiniae. Certainly no one will imagine that what has so long been regarded as the Edwardsian type of structure is not in reality a primary phylogenetic condition, even though we are now obliged to regard the existing Edwardsiae as true hexactinians which secondarily in some cases may represent the more primitive condition.² The facts of embryology speak too strongly regarding the Edwardsian stage to allow of question as to its past occurrence, and I believe that there can be as little question regarding the stages which I have supposed to intervene between the Edwardsiae and the typical Hexactiniae, even though the forms which to-day represent these stages do so possibly only on account of secondary modifications.

¹ *Protanthea* has four imperfect mesenteries which make pairs with the four lateral perfect mesenteries, and twelve others arranged in pairs in the primary exocoels, all being fertile and provided with mesenterial filaments. In addition to these there are, however, as in *Halcurias*, a number of short, narrow mesenteries confined to the upper part of the column and standing in relation to the tentacles, of which there are about ninety-six.

² Compare Van Beneden, Les Anthozoaires in *Ergebnisse der in dem Atlantischen Ocean, etc., ausgeführten Plankton-Expedition der Humboldt-Stiftung*. II. 1898.

It seems inadvisable then to associate *Halcurias* with *Gonactinia* and *Protanthea*, but there still remains a possible association with Hertwig's Antheomorphidae. Unfortunately the forms upon which this family was founded are insufficiently known, but it seems to me that there are reasons for maintaining the position I have already ('98) advocated, that the nearest allies of *Halcurias* at present known are to be found in the family Antheomorphidae. I find myself obliged, however, to recede from the position I held in 1898 as to the distinctness of this family and to return to my earlier opinion, which has received the approval of so critical a taxonomist as Haddon ('98), that the Antheomorphidae should be included in the family Actiniidae, and if this view be accepted it is necessary to refer the genus *Halcurias* to that family also.

This will necessitate no important modification of the definition of the Actiniidae given by Haddon ('98), but as I shall have occasion in the immediate future to consider the family in some detail, I shall postpone a discussion of its delimitation for the present.

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November 10, 1900.

REFERENCES.

- '92 CARLGREN, O. Beiträge zur Kenntniss der Edwardsien. *Öfvers. Kgl. Vet. Akad. Förhandl.* Stockholm. 1892.
- '93 CARLGREN, O. Studien über nordische Aktinien. *Kgl. Svenska Vet. Akad. Handl.* XXV. 1893.
- '97 CARLGREN, O. Zur Mesenterienentwicklung der Aktinien. *Öfvers. Kgl. Vet. Akad. Förhandl.* Stockholm. 1897.
- '99 CARLGREN, O. Zoantharien. *Hamburger Magalhaenische Sammelreise.* 1899.
- '89 DIXON, G. Y. and A. F. Notes on *Bunodes thallia*, *Bunodes verrucosus*, and *Tealia crassicornis*. *Sci. Proc. Roy. Dublin Soc.* N.S. VI. 1889.
- '95 FAUROT, L. Études sur l'anatomie, l'histologie, et le développement des Actinies. Paris. 1895.
- '98 HADDON, A. C. The Actiniaria of Torres Straits. *Sci. Trans. Roy. Dublin Soc.* Series II. VI. 1898.
- '93 McMURRICH, J. P. Report on the Actiniae collected by the U. S. Fish Commission Steamer *Albatross* during the winter of 1887-1888. *Proc. U. S. Nat. Mus.* XVI. 1893.
- '98 McMURRICH, J. P. Report on the Actiniaria collected by the Bahama Expedition of the State University of Iowa, 1893. *Bull. Lab. Nat. Hist. State Univ. of Iowa.* IV. 1898.

OBSERVATIONS
ON THE HABITS AND NATURAL HISTORY OF
AMPHITHOE LONGIMANA SMITH.

SAMUEL J. HOLMES.

IN the present paper I have given the results of my observations made at Woods Holl, Mass., during the past summer on a species of amphipod, *Amphithoe longimana* Smith. Comparatively little is known of the habits of amphipods, and most of what is known has been collected from scattered and casual observations. There is a value in getting together all the facts that can be obtained concerning any one species of animal, so that they may be viewed in their *ensemble* and thus give us some idea of the general life of the creature. For this reason it was deemed best to devote the short time that could be given to the study of amphipod behavior mainly to the observation of a single species.

Throughout the paper I have used many terms which imply the existence in the animal of certain psychical states, such as hunger, fear, and courage, without intending to affirm that such psychical states really exist in the animal's consciousness, or even that the animal possesses consciousness at all. It is difficult to describe the behavior of an animal without the use of terms which have certain psychological connotations. Such terms are here used simply as a matter of convenience in describing actions simply as actions. The Crustacea may or may not be "Reflexmachinen," and Bethe and others may or may not be right in denying that they possess consciousness; but, however this may be, descriptions of actions in psychological terms stand for certain peculiarities of conduct that could not otherwise be easily described, and if the sense in which such terms are used is understood, no confusion need result.

Amphithoe longimana may be obtained in large numbers from the eel pond near the laboratory by simply drawing a

net over the eel-grass. It is abundant during the summer months, a period when most of the other species of amphipods suffer a marked diminution in numbers. It is quite hardy, and may be kept alive for months in small glass dishes, if they are kept covered and the sea water kept fresh by a small piece of *Ulva*. Observations on this species were carried on for nearly three months. Specimens were kept isolated in small dishes and daily observations made and recorded. I was thus able to follow the histories of quite a number of individuals for a considerable period.

Specific Description.

Body slender. Eyes round. Lateral lobes of the head truncated in front. Antennules slender, about as long as the body, the second segment a little longer, but much more slender than the first; third segment from one-third to one-half the length of the second; flagellum much longer than the peduncle. Second antennae shorter than the first, but usually with a longer peduncle; last segment of the peduncle a little longer than the preceding one; flagellum shorter than the two preceding basal joints.

Second, third, and fourth epimera much longer vertically than wide; fifth epimeron about as long as the fourth, but broader and excavated at the upper posterior angle; lower margins of the epimera furnished with very short setae. Postero-lateral angles of the abdominal segments not acute.

First gnathopods in the male elongated, the first joint produced into a rounded lobe at the antero-distal angle; carpus narrow, nearly as long as the hand, and thickly setose on the posterior margin; hand very long and narrow, slightly incurved, and of nearly the same width throughout, although slightly widened near the base; lower margin setose; palm very short, transverse, and rounded at the outer angle; dactyl very large, dentate, and projecting far beyond the palm when closed. Second gnathopods much stouter than the first; first joint with a rounded lobe at the antero-distal angle; hand much broader and stouter than in the first pair; palm oblique, with a deep

sinus near the strongly produced outer angle; dactyl scarcely projecting beyond the palm.

In the female the gnathopods are much shorter and weaker than in the male; the hand in the first pair is less elongated, and the palm is more oblique and more broadly rounded at the outer angle. In the second pair the sinus in the palm is not so deep, and the outer angle not so prominent as in the male.

Peduncle of the first pair of pereopods rather slender, much longer than the rami, and reaching nearly to the tip of the peduncle of

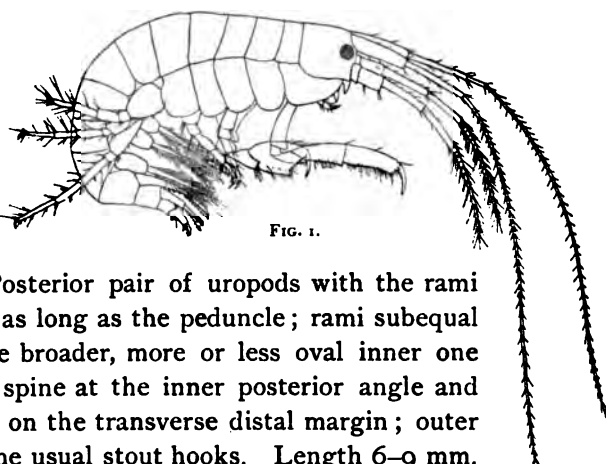
the second pair; inner ramus of the second pair of uropods about as

long as the peduncle. Posterior pair of uropods with the rami scarcely half as long as the peduncle; rami subequal in length, the broader, more or less oval inner one with a short spine at the inner posterior angle and several setae on the transverse distal margin; outer ramus with the usual stout hooks. Length 6-9 mm.

In the older specimens the antennae are relatively more elongated, and the hands of the male relatively longer and narrower. The eyes in the living specimens are red, but become black in specimens preserved in alcohol.

Habitat.

The range of this species as reported by Professor Smith is from Vineyard Sound to New Jersey, and it has been reported from Provincetown, Mass., by Richard Rathbun. It is not uncommon among the seaweed near the shore, and it has been taken at the surface in the vicinity of Woods Holl in the tow net. Its occurrence at the surface is probably due to its having been carried away from the shore by tide currents, as it has a strong tendency to keep among objects of shelter.



Its most favored habitat seems to be the eel-grass, where it finds a convenient substratum upon which to construct its nests. This species is much more common in the eel pond at Woods Holl than outside; the abundance of eel-grass and various algae and the quiet water being conditions which doubtless favor its perpetuation. It is not found on the muddy bottom and does not occur abundantly in the seaweeds near the bottom, but it may be obtained in quantity from the masses of eel-grass at the surface.

Enemies.

In common with most amphipods, *Amphithoe* is doubtless preyed upon by fishes, and it certainly affords one of the principal articles of food of the small but voracious jelly-fish *Gonionemus*. The latter form, however, owing to its unfortunate attractiveness to the zoölogists frequenting Woods Holl, is in danger of not continuing to be a very destructive enemy. It is very common to find *Gonionemus* with *Amphithoe* in its stomach. This crustacean falls an easy victim to its enemy, as it often makes surprisingly little effort to escape, owing possibly to a narcotizing effect of the poison of the nettling organs of its captor. I have seen an *Amphithoe* while swimming vigorously strike against the tentacles of the jelly-fish, suddenly stop, and remain almost perfectly quiet while it was being engulfed.

Food.

Amphithoe lives chiefly upon seaweed. The alimentary canal may usually be seen to contain numerous fragments of red or green algae. Pieces of *Ulva* that are kept in dishes containing these amphipods soon exhibit gnawed margins and, after some time, a marked diminution in size. I have often observed the process of feeding. The *Ulva* is gnawed directly by the mouth parts, without being previously torn away by the gnathopods. The quantity of algae eaten, judging by the amount of excrement voided, is very considerable. In order to ascertain how rapidly the excrement accumulated, a specimen with an abundance of *Ulva* was placed in a clean dish, and

it was found that one hundred and forty-six masses accumulated in twenty-four hours. These masses consisted almost entirely of broken-up cells of *Ulva*, the contents of which had been digested out. By making a very rough estimate based on the size of these masses, it was calculated that the amount of food eaten by the animal in twenty-four hours was about equal to one-tenth of its bulk.

Amphithoe is by no means a strict vegetarian, for it will devour animal food with great eagerness when it can be obtained. It is very fond of bits of flesh of almost any animal, not excluding that of its own species. When aware of the presence of food sufficiently near its nest to be seized without letting go its hold, it will dart out with a quick movement, grab the food with its gnathopods, and suddenly retract itself inside its domicile. When the food is brought in, it is held by the gnathopods while being devoured.

Movements.

Of the movements performed by *Amphithoe*, the beating of the pleopods is the most constant and uniform. Whether the animal is swimming, crawling, or lying quiet, the pleopods are continually engaged in their regular to and fro movement. The motion of these appendages while the animal is at rest serves to create a current of water past the gills in front, and thus aids in respiration. The abdomen, except during swimming, is held strongly flexed, forming a sinus, at the posterior end of which the bases of the pleopods are attached, the tips pointing forward. Small particles suspended in the water may be seen to be drawn in at the sides of this sinus and thrown out at the anterior end, thus indicating the course of the current.

The rhythm of the motion of the pleopods, like the respiratory movements of the higher vertebrates, goes on in a regular way as a rule, but may be checked by impulses from the higher nervous centers. When the animal changes its position, or executes any other decided movement, the pleopods may cease their action for a moment, but soon resume their regular beat

as before. Commonly during swimming the pleopods beat more rapidly, but this is not always the case. When the swimming ceases they drop back into their usual rhythm, whether faster or slower than before. In their motion the three pairs of pleopods act as a unit, keeping perfect time like well-trained oarsmen. If the abdomen be removed from the rest of the body, the pleopods, after a few spasmodic movements due to the shock of the operation, continue to beat rhythmically for several minutes; the three pairs all move with the same rhythm, though somewhat more slowly than before the operation. Moreover, if a single segment with its pair of appendages be isolated, the rhythmic motion of the appendages still goes on for some minutes, but gradually becomes slower and more irregular, until nothing but small twitches indicate the existence of irritability.

When the animal is in a vigorous condition the beat of the pleopods is rapid, but when the creature becomes weakened the beat becomes slower, yet as long as life lasts the pleopods continue their movements. The beat of the pleopods may still persist after the rest of the animal refuses to respond to any sort of stimulation.

The swimming of *Amphithoe* is mainly effected by the pleopods. The first impulse, however, is gained by the sudden extension of the abdomen, which gives the body a rapid forward movement. The abdomen is then held in an extended position and the pleopods, which then hang nearly at right angles to the body, serve to continue the forward motion. When swimming against the force of gravity the motion of the pleopods alone is not sufficient to keep the body going, and when the original impetus becomes exhausted the abdomen is bent forward and again suddenly extended, giving the animal a fresh start. The flexure of the abdomen before every stroke tends to draw the body backward. This, combined with the weight of the animal, causes ground to be lost between every stroke. Swimming towards the surface is therefore accomplished by a series of springs, between each of which the animal falls back more or less. While swimming horizontally the beating of the pleopods is all that is required to keep up the motion; specimens

may be seen swimming about for a considerable time without employing the abdomen.

Amphithoe has a decided disinclination for continuous swimming. Ordinarily it makes only short excursions from one place of concealment to another and generally stops upon meeting with the first solid object that comes in its way, although when situated where there is nothing to which it can lay hold it may swim for some time in a uniform manner. It may swim in various ways, on its side, or with either the dorsal or the ventral surface uppermost, and apparently gets along with about equal facility in any of these positions.

The beat of the pleopods tends to propel the body not in a straight line forward but in a circular course. The pleopods being on the ventral side tend to cause the body to veer around towards the dorsal side. When the body is held somewhat concave on the ventral side, as it often is, this tendency may be balanced or overcome by the tendency to move in circles in the opposite direction. Such a condition is analogous to a person rowing on one side of a boat with the rudder turned toward the side of the oar. By having the body extended to the right degree a straight course may be maintained. The direction of movement is often changed by the animal turning now on one side and now on the other. Circular movements in one direction are thus compensated for by circular movements in another as the animal turns over and a certain general direction of motion is maintained. When swimming on the back a nearly straight course is kept by rolling the body slightly to the one or the other side. Rolling is probably effected by the movements of the hinder pairs of thoracic legs. When the animal is swimming these legs project outward and backward. A downward stroke of these appendages on one side would push the same side of the body upward and roll it over toward the opposite side. In a larger species of amphipod, whose movements are not so exceedingly rapid as those of *Amphithoe*, I was able to see that the rolling of the body was effected in just this way. It is highly improbable that in *Amphithoe* a different method would be employed to produce the same result. However this may be, it is certain that

Amphithoe steers itself while swimming by altering the extension of the abdomen and by rolling from side to side. Lateral bendings of the body could not be seen to play a part in directing the swimming motions, although I have observed this method of steering employed by other amphipods.

Amphithoe longimana, like many other amphipods, is unable to walk over a plane surface. When out of water it is able to make indifferent progress by the characteristically amphipodan gliding movements produced by alternately flexing and extending the abdomen. It is utterly incapable of leaping like the sand fleas and some of their aquatic relatives. Owing to its compressed form, it is unable to maintain itself upright while out of water, or even in water, unless it has some object to which it can lay hold. In its characteristic habitat among the seaweed, *Amphithoe* crawls with considerable agility. The principal organs for crawling are the second antennae, the two pairs of gnathopods, the third and fourth pairs of pereopods, and to a certain extent the abdomen. The antennae are thrown over objects and flexed, thus tending to pull the body upward and forward. The gnathopods are used to seize objects in order to pull the body along. The two following pairs of appendages are employed much as the walking legs of insects, although they move in a nearly vertical plane. The abdomen assists in locomotion by being thrust forward beneath the body until the tip is hooked on to some irregularity of the surface over which the animal is moving when it is extended, thus giving the body a forward impulse. The movement recalls the leaping motion effected by the abdomen in the sand fleas. In fact, very similar motions are performed in both cases, but in *Amphithoe* the motions are much less rapid and energetic. The ambulatory movements of this species are never rapid, owing doubtless to the necessity for keeping the body from falling over on its side. The last three pairs of thoracic legs, although not used directly for locomotion, are indirectly of service in holding the body upright. These appendages, which are bent over the back and have their claws pointing forward, are used to hook on to objects and thus act as organs of support while progression is effected by the other appendages of the body.

Ordinarily *Amphithoe* lies in its nest, with the antennae protruding from the opening at the end. The lower pair of antennae are generally held motionless. The upper pair, however, are usually seen to be moving to and fro, sweeping about in almost every direction. Occasionally their motion is suddenly checked and they are held motionless for a time, but soon their movement is resumed. The significance of these movements will be discussed in a later section. The two pairs of gnathopods are used for a variety of purposes. Occasionally the antennae are bent downward and seized by the gnathopods and the flagella drawn through the space between the dactyl and the palm. The function of this act is probably to strip off any foreign bodies that may become attached to the antennae. The gnathopods are frequently employed to grab passing objects and to reach out and pull in bits of seaweed for the construction of the nest. They are used also for holding the food that the animal is eating and for carrying bits of food to the mouth, where they are taken by the maxillipeds. While not exercising any of their normal functions they may often be seen going through the motions of grasping, with nothing to grasp. This restless activity of the gnathopods seems to be nothing but the exercise of the grasping reflex called forth by some unknown stimulus and having no useful result. The act is performed in all degrees of completeness, from a definite grasping motion to a mere nervous twitch. The mouth parts perform many motions when the animal is not masticating food. These movements, which resemble the normal motions of mastication so far as could be observed, apparently have no functional significance. They take place in specimens kept for a considerable time in dishes in which there is nothing that could serve for food. Like the grasping actions, they are movements which are called forth without the normal exciting cause.

One of the most curious actions which *Amphithoe* performs is its reversal of position in the nest. If the antennae be somewhat roughly struck with a needle, or even if a threatening object suddenly appears close in front, the head and antennae will appear at the other end of the nest. As the nest is a tube

but little wider than the body of its occupant, no one who watches the operation can fail to have a feeling of admiration for the neatness and extreme quickness of this acrobatic feat. The animal executes this "about face" with such rapidity that it is only after watching the operation repeatedly that one can determine how it is effected. As the animal lies in its nest the abdomen is bent forward and the posterior pairs of thoracic legs are directed backward, their claws being usually hooked into the walls of the nest. When about to turn around, the abdomen is thrust forward, its terminal hooks caught in the nest; the tip of the abdomen, therefore, forms a fixed point, and the insertion of the thoracic legs forms another. The contraction of the legs would therefore pull the middle and anterior parts of the body backward. When the head is pulled back some distance, extension of the body occurs, forcing the anterior part of the body through to the other end of the nest. The head end being reversed, the abdomen is loosened and quickly flexed again under the body. The whole operation is completed in less than a second, and the animal may be made to repeat the performance several times in rapid succession.

Nests and Nest-Building.

The nests of *Amphithoe* are tubular structures which generally exceed somewhat the length of the animal. They are made of a web-like material which, under the microscope, may be seen to be a network of exceedingly fine threads. The nests are usually constructed among the branches of the red seaweeds or upon the leaves of eel-grass or the fronds of *Ulva*. When built upon *Ulva* the nest is generally located in a wrinkle or fold of the surface which affords a partial shelter. The nest is open at both ends and is of about the same diameter throughout. Foreign materials, such as bits of seaweed, are usually woven into the nest and greatly add to its efficiency as a means of concealment. *Amphithoe* frequently leaves its nest, but I could find no evidence that it would return to its own nest more readily than to any other; it will simply enter the first unoccupied nest that comes in its way. When established in

a nest *Amphithoe* is driven out only with difficulty. A member of its own species that approaches is grabbed at and usually driven off, and the creature appears to be on the alert to keep out all intruders. The approach of a more formidable-looking object causes the animal to retreat farther back into its nest. If the antennae are stroked with a needle, a sudden somersault will be executed and the head will appear at the other end of the nest. Then it usually requires quite a series of pokes to make the creature quit the nest entirely. The instinct to remain in the nest when danger threatens presents a marked contrast to the quickness with which flight is made when the animal is roaming free.

A new nest is constructed in a remarkably short time, often in less than a half hour. If a few specimens be placed in a dish of sea water containing a little seaweed, nests will be woven on the seaweed and on the lower surface of the dish, and in a short time the number of nests may greatly exceed the number of specimens. Those localities are chosen which give the animal a maximum of contact with solid objects. In dishes in which specimens were kept I have nearly always found several nests along the angle between the bottom and sides, although the seaweed kept in the dishes afforded localities better adapted for concealment. The choice of a spot for a nest is apparently largely a matter of thigmotaxis. When the animal remains in a spot for some time, the nest-building activities begin, and where contact with different sides of the body is secured, as between the branches of seaweed, in the wrinkle of an *Ulva* frond, or in the angles of a glass dish, it remains quiet. If *Amphithoe* is observed while constructing its nest, the first and second pairs of pereopods will be seen to be busily engaged in moving back and forth from point to point along the surface on which the web is being laid down. The first and second pereopods contain large glands which are connected with a duct which opens at the tip of the claw. The material for the web is secreted by these glands and probably hardens soon after its emergence, like the web of a spider. A very fine thread of web may frequently be seen passing out from the small opening at the tip of the claw. As the tip of

the claw touches one point after another, the web, as it is drawn out, is fastened to different places. By moving back and forth and rolling around during the weaving process, the animal constructs its tubular dwelling. Several specimens from which I clipped the claws from the first two pairs of pereopods were kept for several days and did not construct a single nest.

During the construction of the nest, *Amphithoe* will reach out and draw in bits of algae and other objects that lie near and incorporate them into its dwelling. In a few cases I have seen long pieces of algae bitten in two and used for this purpose. As *Amphithoe* lives largely on algae, this biting may not have had any special reference to nest-building, but may have been a manifestation of the ordinary reaction to food. In *Microdeutopus*, Smith has observed that the excrement of the animal is worked into the web; but in *Amphithoe*, whose nest-building habits seem to be very similar, no such process could be observed. The excrement is passed out of the nest, accumulations of it usually being observable near the two ends.

Moulting.

Amphithoe was found to shed its skin more often than was anticipated. Most of the specimens I kept isolated as long as a week moulted once, and out of four specimens in which I have records of the dates of two successive moults of the same individual the interval between moults in three cases was seven days, and in the other case eight days. These specimens were of the usual size. How rapidly moults occur in different periods of the life history of this species I cannot say.

The process of moulting in *Amphithoe* occurs in the same manner as has been described in other species of amphipods. The skin splits transversely along the line joining the head and thorax, and on either side of the thorax is a longitudinal split which occurs between the upper margins of the epimera and the lower margins of the thoracic rings. This split may extend along all the thoracic segments. The head and antennae are pulled backward out of their investment and the posterior part of the body is pulled out forwards, the old skin, after being

shed, remaining intact except at the lines just mentioned. The moulting process takes several minutes at least and is accompanied by considerable muscular effort to get out of the old skin.

In the several cases in which I observed the process, *Amphithoe* leaves its nest to divest itself of its skin, and I have never observed a moult in a nest but always some distance away. After moulting the animal is rather quiet and cannot easily be enticed from its nest by food. I have observed several cases in which death occurred during the moulting process. In one case moulting was not completed for several days. The specimen was observed August 17 with the head and tail ends drawn partly out of the old case. The next day the head and antennae were still not completely drawn out, but the rest of the skin was kicked off. On August 21 it was still in the same condition, the feeble beating of the pleopods giving evidence of failing strength. On the next day it died, the head and antennae still only partly extricated from their old covering. In several cases the antennae were observed to become broken off in the process of moulting, but I have seen no cases in which other appendages became lost in this way. The antennae are the appendages most liable to more or less complete loss from other causes, but owing to the rapidity with which these organs can regenerate this loss can produce only a temporary inconvenience. The cast-off skins are found sometimes on the bottom, and often floating on the surface of the water, and in a short time after they are shed become filled with swarms of protozoa.

The Seat of Smell.

Much has been written concerning the seat of the olfactory sense in the Crustacea, but most opinions on the subject have been based on morphological instead of experimental evidence. The work of May and Bethe affords good evidence that in the decapod Crustacea the seat of the olfactory sense, or, as Bethe prefers to call it, of chemoreception, is in the first antennae, as analogy with the insects would lead one to suspect. The first antennae are not, however, according to Bethe, the only seat

of chemoreception. In *Carcinus* the removal of the first antennae as far as the first basal segment is followed by a marked diminution of the power of reaction to chemical substances in the water. A *Carcinus* when the eyes are blackened over will find pieces of food when placed at some distance, by the sense of smell. When the first antennae are removed at the first basal segment, Bethe found that food may be placed as near as 10 cm. to the animal without calling forth any efforts to obtain it. When the food is brought close to the mouth or close behind the animal without contact with the body, it is seized and eaten. The first antennae, therefore, while they may be the main, are not the sole source of the reception of olfactory stimuli.

My own observations on *Amphithoe* led me, before becoming acquainted with Bethe's results, to infer a double seat of the sense of smell. In *Amphithoe*, as in *Carcinus*, the first antennae seem to be the most important olfactory organs. While the animal is at rest in its nest the antennules are kept swaying to and fro in different directions, as if they were being employed to explore the surroundings. If a small bit of flesh is held on a needle or in a fine pair of pinchers and carefully brought near the animal, the antennae check their random movements and make one or more strokes in the direction of the bit of flesh; often the antennae are held for some time in the direction of the object. On bringing the flesh nearer, the animal may be seen to adjust itself in the nest for a sudden spring, and if the flesh is sufficiently near to be touched by the antennules the amphipod makes a sudden dart from the nest, seizes the object, and draws quickly back again, never letting go its hold, however, of the nest. The animal as a rule readily distinguishes between the contact of flesh and that of a body not serviceable for food. Only rarely does touching the antennule with a needle call the animal forth from the nest. It may be deceived more often if, when excited by the presence of meat near by, one of its antennules be touched with a needle; then it may dart out towards the needle and even seize it. But the animal responds much more surely, as I have found by repeated experiments, when the antennules come in

contact with the food itself, even when the animal is excited by the presence of food in its vicinity. The darting forth, therefore, is apparently caused, not merely by a tactile stimulation, but by a chemical stimulus from the food. The antennae are delicate tactile organs, and tactile stimuli may assist in calling forth the actions which result in the seizing of food, but tactile stimulation alone generally fails to accomplish this result.

After *Amphithoe* has made a meal of fleshy diet it becomes quite indifferent to the presence of that kind of food in its vicinity and no longer darts forth to grab bits of flesh brought in contact with its antennules. Different individuals present very different degrees of eagerness for animal food, owing doubtless to varying intervals of time since their last repast.

Sight has probably little to do with the food reactions of the animal. When the head is completely withdrawn in the nest the animals often give signs of perceiving food and dart after it when brought in contact with the antennules. In many cases the nest is so opaque that the animal cannot see through it with any distinctness, and under these circumstances, when the head was entirely withdrawn into the nest, I have often brought bits of meat so they would be touched by the antennules only when they were strongly bent backwards. Although the meat was out of sight, the amphipod would dart out, bend backwards, and seize the morsel. If the desired object is out of reach of the antennules, the amphipod will not spring for it, although it may be seen to make ready to do so. It will not go to the length of leaving the nest to seize food, even if its conduct betrays evidence of keen hunger. An object near enough to be struck by the swaying of the antennules is sufficiently near to be seized by the animal without letting go its hold of the nest. It is a noticeable feature of the species of *Amphithoe* and related genera that the antennules are, roughly speaking, of about the length of the body. This feature is not improbably correlated with the similar tube-dwelling habits of these forms, the length of the antennules gauging the length of a safe and successful spring.

The effect of removing the antennules of *Amphithoe* is greatly to lessen responsiveness to olfactory stimuli. The

shock of the operation has a very temporary effect, for in a few hours the animals behave with their usual activity. Meat brought in contact with the second antennae is generally not seized. This, however, is not always the case, for in several instances I have found that contact with the second antennae causes the grasping reflex. I was inclined at first to attribute a certain olfactory sensibility to the second antennae, but I found later that the animal reacts about as well to olfactory stimuli when both antennae are removed as when the second alone remain. In specimens with both antennae removed near the base, leaving only the first joint of the peduncle of each pair, there was no reaction to food placed in what would have been within easy reach of the antennae before their removal. If a piece of meat is placed about 2 mm. from the mouth of the amphipod, it is generally allowed to remain untouched for several seconds and then suddenly seized and eaten. The morsel is seized by the gnathopods and at the same time bitten at with the mouth parts. The reaction is not an immediate one, such as is brought about by contact of the antennules with food. It appears to be necessary for the food to remain awhile in close proximity to the animal before its edible nature is perceived; when this occurs the seizing takes place quickly enough. I have tried to induce the animals to take bits of substance of the general appearance of fragments of meat and brought very close to the mouth parts, but they are apparently able to distinguish, before any contact with the object occurs, whether or not it is of an edible nature. It is true that *Amphithoe* often grasps objects that lie near by, pulls them back, and incorporates them into the structure of its nest, and it might be inferred that the seizing of meat lying close to its mouth by a specimen with both antennae removed is an expression of the nest-building instinct to seize any small object within reach for building material. The reactions in the two cases, which I have observed many times, differ. An object used for the construction of the nest is reached for and pulled back to the nest and not as a rule brought in contact with the mouth. A bit of meat is grabbed at and bitten at in the same act. This difference in reaction and the fact that

the animals seize bits of meat when they cannot be induced to pay attention to other objects of similar appearance convinced me that the reaction to food was caused by chemical stimulation from diffusing substances in the water.

Removal of the second pair of antennae, the first being left intact, was not found to exert any marked influence upon reactions to chemical stimuli. The second antennae may transmit olfactory stimuli; it would be difficult to prove they do not in a certain degree, but the evidence obtained does not justify us in attributing to them this function. When, after removal of the first antennae, *Amphithoe* responds when food is brought in contact with the second antennae, the reaction may be due to the animal becoming aware of the presence of food through some other organ, contact with the antennae indicating that the food is sufficiently near to be seized. In other words, the reaction may be due to purely tactile stimulation, the animal being keyed to this reaction by the excitement of olfactory stimuli from some other organ. What other organ, or organs, may serve to transmit olfactory stimuli is uncertain. This has not been determined in the decapod Crustacea, which afford the only other instance in which a double seat of the olfactory sense has been suspected, or in fact in which, so far as I am aware, any experimental evidence has been adduced as to any location of this function at all. It seems probable that some of the mouth parts have some olfactory function, as they afford the most obviously appropriate location for such a sense. Owing to its small size, *Amphithoe* is not a favorable form in which to decide this question, and the attempt to do so was not made.

Color and Color Changes.

One cannot but be struck, when examining a number of specimens of this species, with the marked differences in color presented by different individuals. Some are bright green, like the bright green seaweeds; others may be nearly colorless; a few are of a light blue green tint, and many range from a light to a dark reddish-brown. The same individual may take on, at different times, all these varieties of coloration. The color

differences are produced by the variation of five elements: (1), the color of the chitinous integument; (2), the color of the blood and tissues; (3), the contents of the alimentary canal; (4), the color of the sex glands; (5), the pigment cells. The first of these factors is, perhaps, the least important and is not subject to great variation. The exoskeleton over most of the surface of the body is colorless; on the antennae it is marked with transverse reddish-brown bands which give the light reddish-brown annulations of these organs. This color is seen as distinctly in the shed skin as in the living animal.

The color of the tissues and blood is subject to great variation. The green color of *Amphithoe*, or the blue green tint when it occurs, is due to some coloring matter that is uniformly diffused throughout the animal. In some specimens there is a sufficient amount present to give the animal a brilliant emerald green, but many may be found in which not the slightest trace of green coloration could be detected. This green color may be seen to undergo marked changes in intensity if individuals be watched for several days. The blue color is much rarer. One specimen in which this blue coloration was strongly marked was kept under observation for several days. After five days most of the blue color had disappeared, the green becoming more nearly like the typical green of other forms. After six days the green was not to be distinguished from the ordinary type; the green color then gradually became fainter, and on the ninth day the tissues were whitish, scarcely a trace of green being visible. During all this time the specimen ate abundantly of green algae, judging from the amount of excrement consisting of *Ulva* cells that accumulated in the dish.

The contents of the alimentary canal influence to a considerable extent the general color effect produced by the animal when seen by the naked eye. If they consist largely of green *Ulva*, they tend to give the animal a greenish appearance. If the *Ulva* has been subjected some time to the action of the digestive juices and become a yellowish color, it tends to give the animal a corresponding yellowish aspect. Light becoming colored by passing through the alimentary canal is reflected and re-reflected in the tissues and tends to make them appear

a corresponding color. I have several times cut off parts of the body to see if their color might not have been due to this cause; but whatever effect this factor may have it is certain that the green color of the blood and tissues is not entirely caused in this way as it may easily be observed in the isolated appendages. The part played by the sexual glands in the coloration of this species varies greatly owing to the variation in the size of these organs.

It is to the pigment cells that the most marked changes of color are due. These cells are of two kinds, — reddish-brown pigment cells, and cells with a pale green pigment. The latter play an insignificant part in the coloration of the animal, as they are pale in color and few in number, there being often not more than a dozen on the entire surface of the body. The pale green color appears most clearly in transmitted light; in reflected light they are of a silvery hue. Their size is about the same as the largest cells with red pigment. Like the latter, they are very richly branched, but were not seen to undergo much variation in the distribution of their pigment. They are mainly confined to the epimera, being usually situated near the lower margin.

The most important elements in determining the color changes are the reddish-brown pigment spots. These spots are scattered all over the body and are found also on most of the appendages, especially towards the proximal end. When extended the pigment spots are large and very richly branched, forming most beautiful objects when seen under the microscope. When fully contracted these spots assume the form of round dots, and all stages of expansion may be seen in different specimens, or even in the same specimen, between the most contracted and the most expanded state. There are a few large spots near the lower edges of the epimera that are generally found in an expanded condition. Even when most of the spots over the surface of the body are contracted, these few spots, which may be not more than a dozen on each side, are usually conspicuously large. This circumstance affords a convenient means of distinguishing *Amphithoe longimana* at a glance from other species of the same genus.

The pigment spots of *Amphithoe* apparently contain but one kind of pigment. The whole system of chromatophores is much less complicated than that which Gamble and Keeble¹ found in *Hippolyte varians*, and the power of sympathetic color changes in relation to surrounding objects much less perfectly developed. *Amphithoe* may be said to adapt its color to its environment, but in only a rather rough way compared with the remarkable protective color changes of *Hippolyte*. Specimens taken from the eel-grass are very apt to be of a greenish color, and specimens taken from among masses of red seaweed are usually colored somewhat like their environment. While on the eel-grass, they are usually exposed to the light and the pigment spots are in a contracted condition. This allows the green color of the tissues to be seen, and the animal has, consequently, a greenish aspect. In the masses of red seaweed the animal is usually more shaded and the pigment spots become expanded, giving the animal a reddish tint which helps to conceal it in its environment. Moreover the alimentary canal in specimens found in the red seaweed often contains a greater or less quantity of this alga, and this also helps to color the animal in a protective manner. So far as the green color of the tissues is concerned there appears to be no difference between the animals from different habitats. The change from green to reddish, or the reverse, completes the range of adaptive color variations in this species so far as they are induced by changes in the environment. And this kind of color change is the one best adapted to afford protection to the species in its usual habitat.

The pigment spots of *Amphithoe* change very slowly; it generally requires some hours to effect a change from the expanded to the contracted condition. It was found by several experiments that exposure to bright light causes the pigment spots to contract, while specimens that have been kept in the dark for several hours generally have the pigment spots much expanded.

¹ *Quart. Journ. Micr. Sci.*, 1900.

Sexual Habits.

Concerning the sexual habits of *Amphithoe* I have little to add beyond what is known among other amphipods. The male carries the female about for a considerable period and maintains his hold against efforts to dislodge him with great pertinacity. The instinct to retain hold of the female is sufficient to overcome all fear, and it is difficult to separate the male without injury. The posterior part of the body may be cut off, and yet the anterior portion retains its hold of the female as long as sufficient vitality remains. Ordinarily the male retains his hold of the female by hooking the claws of his pereopods beneath the edges of the epimera. The gnathopods are not generally employed for this purpose, but are called into use when a sudden disturbance renders the hold of the male insecure. The female remains remarkably passive when carried about by the male. Her body is usually held quite strongly flexed and the male does the swimming for both, the female being transported as so much dead weight. While carried by the male the female seems much less responsive to stimuli than when free. When poked by a needle she often makes little motion, but the more alert male is generally aware of the disturbance and carries her away from the seat of annoyance.

The Disposal of Excrement.

In its natural position *Amphithoe* lies so that the excrement that is voided would be deposited in the nest. Yet the excrement is never found in the nest but at some distance from either end. At first I supposed that it was carried out by the current of water produced by the movement of the pleopods, but, after watching the animal, I noticed that when excrement was extruded the abdomen was bent forward and the gnathopods reached back and seized the mass as it was ejected from the intestine, and passed it out of the front end of the nest. This act was observed three or four times, but whether it is always performed when excrement is extruded I cannot state. In one individual lying outside a nest on the bottom of a glass

dish I noticed that when excrement was passed the abdomen was bent forward and the mass seized by the gnathopods and passed forward, just as it is when the animal is in the nest. There was of course no use in seizing the excrement under the circumstances, but the act was performed in the usual instinctive way nevertheless.

Timidity and Pugnacity.

Specimens of *Amphithoe* are very ready to attack other amphipods that come near and drive them away. The animals appear to be on the alert to prevent any other individual from gaining access to the nest, — so much so that they very frequently spring out at a passing amphipod and bite at it in what appears to be a particularly vicious and hateful manner. The individual attacked does not, so far as I have observed, attempt any defense but precipitately flees from the spot. Anything that properly could be called a fight is never engaged in; a passing nip with the gnathopods, or bite with the jaws, is all that seems to occur in the nature of hostilities.

While ready to dispute the entrance of another amphipod from in front, *Amphithoe* generally quickly flees from its nest when an intruder enters from behind. One often sees the occupants of nests routed out by others entering in this way, and I have seen one individual that was expelled by another entering behind it swim around, enter the other end of the nest, and drive out the intruder. Courage in *Amphithoe* depends in great measure on whether the attack is made from in front or behind.

Outside the nest *Amphithoe* is very timid. It does not attack its fellows except by giving an occasional nip when accidentally colliding with them, and it flees quickly when disturbed. It can very rarely be induced to seize meat, however hungry it may be, and however carefully the food be presented. Even when in the nest, care has to be taken in offering the animal food lest it be alarmed by one's movements. This alarm is manifested by withdrawing a short distance in the nest. When an animal to which meat is presented withdraws in this way, I find that it is useless to attempt to

induce it to take food for several minutes, until its fright wears away. Experience in feeding these animals soon enables one to tell whether or not they have become frightened by your actions.

So far as my observations go they indicate that *Amphithoe* has very little true pugnacity. It does not engage in a conflict in order to overcome an adversary, as many decapod Crustaceans do; it fights only in self-defense. The attacks on other amphipods passing by the nest are simply measures to keep out unwelcome visitors. Had not these forms the instinct to keep the nest to themselves, several individuals would often crowd into the same nest much to their mutual inconvenience.

Phototaxis.

Amphithoe, like most of the aquatic Gammaridea, is negatively phototactic. The specimens experimented with were placed in an elongated, rectangular dish contained in a box open at one end and above and blackened on the inside. When placed near a window, either in direct sunlight or so that rays of diffuse daylight fell obliquely into the dish, the animals would swim towards the end of the dish farthest from the source of light. When the dish was turned about, they swam back again to the other end. In lamplight they may be driven alternately from one end to the other by moving the lamp back and forth to opposite ends of the dish. The endeavor was made to make *Amphithoe* positively phototactic by altering the temperature and concentration of the sea water, but only negative results were obtained. The animals remain negative until the water is heated to about 90° Fahr., when little responsiveness to light remains. Further heating causes heat rigor to supervene. Increase of temperature may be carried to the point of producing death without changing the direction of the phototactic movements. It is not probable that cooling the water below the normal temperature would alter the phototactic response. Decrease of temperature, other things equal, tends to give rise to negative phototaxis in *Orchestia*, and increase of temperature has the effect of making this form more positive.

Raising the temperature has the effect of making *Gammarus mucronatus* positively phototactic, but an increase of only a few degrees beyond the point where its phototaxis changes produces heat rigor. Possibly *Amphithoe* might be rendered positively phototactic could it endure a somewhat higher temperature.

Neither increasing nor decreasing the concentration of the sea water was found to effect a change in the direction of phototaxis. A more extended account of phototaxis in the Amphipoda will appear in another paper.

Thigmotaxis.

The tendency of *Amphithoe* to keep in contact with solid objects is one of the most conspicuous features of its behavior. This tendency is apparently one of the fundamental instincts of the group, as it is exhibited very strongly by most of the Amphipoda. When placed among branched seaweeds, *Amphithoe* stops only after it works its way among the branches where there is contact on several sides of the body. In *Ulva* it comes to rest in a fold of the frond. When placed in a glass dish containing nothing but sea water, it swims about restlessly and eventually comes to lie quiet in the angle between the bottom and side. The instinct to crawl into an empty nest is an expression of the same tendency. Were it not for its thigmotaxis, the whole conduct of the animal would be very different from what it is. Many other instincts, like the nest-building instinct and the instincts associated with it, are built upon this fundamental reaction as a foundation. It does not seem improbable that the instinct of the female to remain perfectly quiet while carried about by the male, and even the strong propensity of the male to seize and retain hold of the female, may be but modified and specialized forms of thigmotaxis. Given variations of responsiveness to contact in different parts of the body, and variations in the manner in which the responsiveness is exhibited, we would have the means by which thigmotaxis might be modified by natural selection into more specialized forms of behavior. If the origin of the various

forms of amphipod behavior could be traced, it would be found, I believe, that thigmotaxis is the mother of many instincts.

The Instincts of the Young.

When the young of *Amphithoe* quit the maternal brood pouch, they have a quite different appearance from the adult. They are whitish in color, with only a very few pigment spots, located for the most part on the epimera, each epimeron containing often but one spot. These pigment cells are of a greenish-gray color and apparently have none of the reddish-brown pigment which is found in those of the adult. By reflected light they have a light greenish-silvery appearance much like the large light green cells of the adult, but they are very much smaller and much less branched. The head is relatively large, but the eyes are small and red and composed of only six ocelli, one of which is in the center surrounded by five others. The number of ocelli, therefore, increases very greatly as the animal grows older. Both pairs of antennae are short, the flagella of the first pair consisting of four joints and those of the second pair of but three.

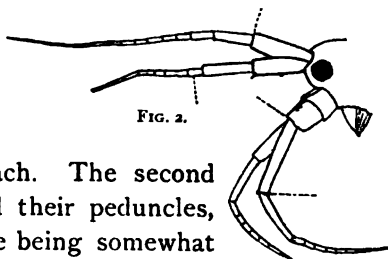
The young when first hatched are in a feeble condition and are carried about for a few days in the brood chamber of the mother. Before they are hatched one can easily see the beating of the heart and the peristaltic movements of the intestine. Shortly before their emergence the young may be observed flexing and extending the body in the effort to break the shell. In one case I removed a lot of eggs from the brood pouch of the mother while they were hatching. Those that had not yet emerged were very vigorous in their movements within the shell. When hatched they were unable to swim, and their movements were irregular and little cöordinated. The next day a few had died; the others could swim feebly, but none well, and if they became caught in the film at the surface of the water they were unable to overcome the surface tension and get free. In another case I removed from the brood pouch of a female several young that had been hatched only for a short time, as they were feeble and scarcely able to swim.

The swimmerets, however, beat rapidly, and the maxillipeds and gnathopods were in constant motion. The antennules were moved more than the antennae, but their movements were more jerky and irregular than in the adult. The next day several others came out of the brood pouch of the mother of their own accord. They were a little more active than those that had been removed the day before and exhibited apparently a faint negative phototaxis. They had been out only a short time when they began constructing nests. These nests were the same in shape as those formed by the adult, and the behavior of the young in relation to the nest was almost exactly like that of the older individuals. One of these young impaled on a needle was presented to one of the same brood lying in its nest. At first the animal gave signs of timidity and withdrew further into its nest. After some waiting the animal emerged a little and began waving its antennules in the usual manner. When they touched the food the creature darted out quickly, seized it, dragged it back, and proceeded to devour it at leisure. Apparently, it is only one or two days after hatching that the young get effective control of their movements, and they probably remain at least that long in the maternal brood pouch. When they are sufficiently active to make their exit they are equipped for the business of life. It was a matter of some surprise to observe how perfectly endowed the young are with the instincts of the parent forms. Their behavior in almost every respect seems exactly like that of the adults. The nest-building, movements within the nest, such as waving the antennules, retraction, reversal of position, springing out after food, jumping after passers-by, signs of timidity, as well as the general behavior outside the nest, are all carried on just as in individuals many times their size.

Regeneration.

No attempt was made to study the power of regeneration in *Amphithoe*, but it may be worth while to record a few observations that were made incidentally on the regeneration of the antennae. The antennae were removed in several specimens

while studying the reactions of the animal to olfactory stimuli, and it was noticed that the regeneration of these appendages took place with considerable rapidity. Several observations were also made on specimens in which the antennae were found to have been accidentally lost. In a specimen (Fig. 2) observed August 13 the left antennule had been removed at the end of the second joint, and the right one at the end of the first. Of the second antennae the left member was off at the end of the fourth joint, which had a small, apparently regenerated knob at the end, and the right was gone at the end of the third and had a black tip. On August 16 the specimen moulted and the antennae appeared as follows: The right antennule had regenerated the last two joints of the peduncle and a flagellum of twelve joints; the left antennule also had completed its peduncle and regenerated a flagellum of seven joints, two of which were partially constricted in the middle and may have later become



separated into two joints each. The second antennae had both completed their peduncles, the fifth joint in the right one being somewhat longer than in the left. The left antenna as a whole, however, was longer than the right, and its flagellum consisted of nine joints, while that of the right was composed of seven. How long the antennae had been injured when the observation was first made I cannot state, but it was certainly since the previous moult, so these appendages were probably regenerated in not much over a week and possibly in a less time. In another specimen first observed August 13 the left antennule was gone at the end of the second joint, and the right one at the middle of the second joint. The left second antenna was off near the end of the fourth joint, and the right one at the end of the third. After moulting, which occurred on August 14 or 15, the right antennule had a completed peduncle and a flagellum of six joints; the left antennule did not regenerate. Each of the second antennae had a complete peduncle, and the left one had a flagellum

of three joints and a minute terminal fourth joint; the right one had a flagellum of four joints.

The Effect of Cutting the Animal in Two.

It has been found in many of the lower animals that, after removal of the posterior part of the body, the anterior portion manifests little signs of pain and acts as if no injury had been received. Many interesting cases of this kind have been collected by Dr. Norman in his paper entitled "Do the Reactions of the Lower Animals against Injury indicate Pain Sensations?"¹

In order to see how the removal of the posterior part of the body affects the behavior of *Amphithoe*, I cut the animal in two just behind the third thoracic segment. In the posterior piece the thoracic legs moved but little, but the pleopods kept up their rhythmical beating for fully a half hour. The anterior part of the animal apparently suffered little discomfort, since, after the operation, it behaved much as if forming a part of the whole organism. The animal lay moving its antennules to and fro and making the usual movements of its gnathopods, just as an uninjured specimen would do. A piece of meat was brought near so that it was struck by the movements of the antennules; it was first swept a little nearer by the second antennae, and then quickly grabbed by the gnathopods, drawn in, and eaten, although the food could pass through only the small part of the alimentary canal that remained. Since this reaction in the normal animal is readily prevented by fear, and only occurs when the animal is hungry, it is somewhat surprising to find it performed after such a serious injury as the loss of the posterior half or more of the body. The animal behaves in such an apparently normal manner after this operation that it would seem as if the loss was scarcely felt. Owing to the loss of the posterior part of the body, many of the actions of the animal are naturally impeded or prevented. It cannot, for instance, get meat except when it is placed quite near, as it is unable to make its accustomed spring out of the nest. But in general,

¹ *Am. Journ. Phys.* Vol. iii, p. 271. 1900.

so far as the actions of the animal are modified, they are affected rather by the loss of certain organs than by any shock to the central nervous system. The loss of blood consequent upon the operation soon leads to weakness and finally death. Were it not for the profuse bleeding that occurs, the separated halves of the body could probably be kept alive for a long period.

UNIVERSITY OF MICHIGAN,
ANN ARBOR, MICH.

BIOLOGICAL BULLETIN.

NOTES ON THE HABITS OF PYCNOGONIDS.¹

LEON J. COLE.

THE Pycnogonids constitute a small and well-defined group, especially interesting on account of their peculiar and unique structure. In general they have been largely neglected by naturalists, especially as compared with some other groups, but they have received considerable attention from specialists, and several excellent monographs have appeared dealing with their structure and classification; in fact, all the literature of the group is very largely systematic or strictly morphological. The embryology has been worked out for some forms by Dohrn and Hoek, and in this country by Morgan,² who has also given an extended account of the metamorphosis of *Tanystylum*, and considerations on the phylogenetic position of the Pycnogonida.³ The principal systematic work in this country has been done by Wilson,⁴ who described some fifteen species found on the New England coast. Practically nothing has been written on their habits, which ought, it would seem from the isolated position of these animals, to offer some very interesting comparisons with those of other arthropods. An excellent opportunity for this work was offered during the past summer at the

¹ From the Zoölogical Laboratory, University of Michigan.

² Morgan, T. H., "A Contribution to the Embryology and Phylogeny of the Pycnogonids," *Stud. Biol. Lab. J. Hopkins Univ.* V., No. 1, pp. 1-76. 1891.

³ Morgan, T. H., *loc. cit.*, and "The Relationships of the Sea-Spiders," *Biol. Lect. Marine Biol. Lab.* for 1890. Seventh Lecture, pp. 142-167. 1891.

⁴ Wilson, E. B. (a) "A Synopsis of the Pycnogonida of New England," *Trans. Conn. Acad.* V., pp. 1-26. 1878. (b) "The Pycnogonida of New England and Adjacent Waters," *U. S. Fish Com. Rept.* for 1878, pp. 463-506. 1880.

Marine Biological Laboratory at Woods Holl, and at the suggestion of Dr. S. J. Holmes, to whom I am also indebted for much help and advice, I undertook to ascertain something of the habits and reactions of the forms found there. The present paper embodies some of these observations, which though far from complete seem to be of interest. I hope to be able later to supplement them by a more comprehensive account of the biology of the group. Much of this work is rendered difficult by the fact that the animals are not easy to observe under natural conditions.

As has been noted by Morgan and other authors, there are three species of Pycnogonids to be found at Woods Holl, representing as many genera. These peculiar animals may be found in nearly every collection of hydroids made from the piles or dredged up from the bottom, their long legs appearing to be hopelessly tangled among the stems of the hydroid as they kick slowly about in an aimless but persistent fashion. Perhaps the commonest of these, and by considerable the largest (extent 40 mm. to 50 mm.), is the dark purple colored *Anoplodactylus lentus* Wilson (= *Phoxichilidium maxillare* of Morgan and others), which is especially abundant in colonies of Eudendrium taken from the piles. A smaller species of a yellowish color, *Tanystylum orbiculare* Wilson, measuring about 7 mm. in extent, was found fairly abundant in a yellowish hydroid almost the exact color of the Pycnogonid. *Anoplodactylus* was on no occasion found among the light-colored hydroid, nor did I ever find a specimen of *Tanystylum* among the dark colonies of Eudendrium, where *Anoplodactylus* is fairly inconspicuous, though I have found the latter among the much lighter colored *Bugula* growing near the Eudendrium. I am not prepared to say that this is a case of color adaptation, as my observations were too limited to confirm this view, but merely throw out the suggestion for what it is worth. And it is worth remarking in this connection that the third representative occurring in this locality, *Pallene brevirostris* Johnston (= *P. empusa* Wilson), which is a slender whitish or more or less transparent form and is very hard to see for this reason, was found to be much more generally distributed than either of the other

species, being found among both light and dark colored hydroids and algae. Pallene impresses one as the smallest form of the three owing to its extreme slenderness, though it is really almost twice the extent of Tanystylum, measuring some 12 mm. to 13 mm. across.

Swimming and Crawling Movements.

The activities of the three species of Pycnogonids under consideration are in a way directly correlated with their structures. Tanystylum, a short-legged and compact form, is very sluggish and inert; if placed at the surface of a dish of water, it kicks hardly at all, but sinks immediately to the bottom,¹ where it does not attempt to crawl but usually draws its legs together over its back and remains quiet. Pallene, on the other hand, under the same conditions does not sink to the bottom, but by vigorous kicking movements of its long slender legs remains suspended in the water, for a considerable time at least, its further movements being determined by the conditions, one of the most important of which, as will be shown later, is light. In the actions of Anoplodactylus there is great individual variation, but in general it may be said that they are intermediate between those of Tanystylum and Pallene. Some specimens sink almost at once to the bottom, where they rest in whatever position they may strike; others may crawl along upon the sand, or partly swim, touching the sand with only the tips of certain of the legs; or still others may swim entirely free from the bottom. As with Pallene, just what the animal does appears to depend largely upon the conditions. Most of my observations were made upon Anoplodactylus, for the reason that it was easiest to obtain and of convenient size for observing the movements in detail.

Before going further it may be well to give a brief explanation of the terminology which I shall use. Various authors have used different names for the seven pairs of appendages

¹ In these experiments the bottom of the dish was covered by a layer of fine sand. The depth of water was usually about 5 cm. to 7 cm., though deeper water was tried with no difference in the results.

of the Pycnogonids, largely as they regarded them homologous to the appendages of the Crustacea or to those of the Arachnids. Dohrn obviates this difficulty by simply numbering them in their natural order, I-VII (Fig. 1).¹ For convenience I shall speak of the third pair as the ovigerous legs (these are absent in the female of *Anoplodactylus*), and of pairs IV-VII simply as the first, second, third, and fourth pairs of legs

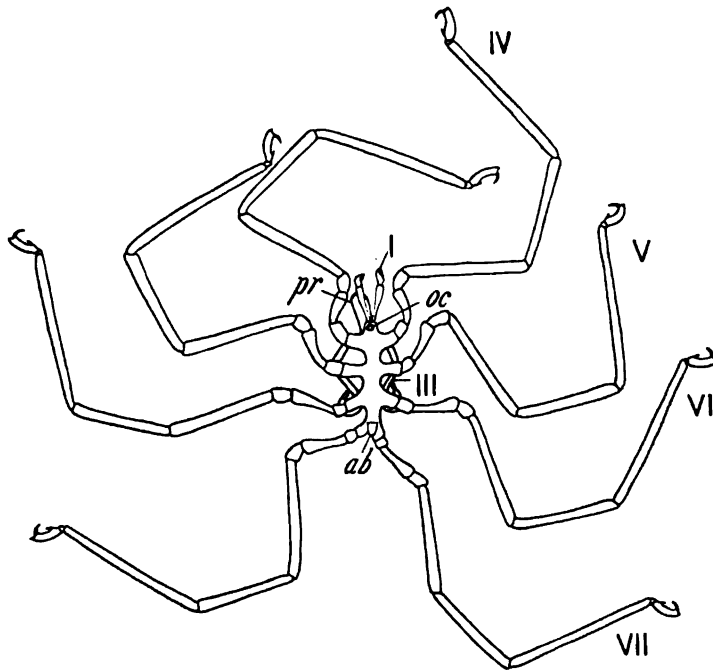


FIG. 1. — Male *Anoplodactylus lentus*, dorsal aspect: *ab.*, abdomen; *oc.*, oculiferous tubercle; *pr.*, proboscis; 1, chelifori; III, ovigerous legs; IV-VII, walking legs. $\times 3$.

respectively, or, to distinguish them from the ovigerous legs, as the walking legs. Each of the walking legs is composed of nine joints (including the terminal claw), and all four pairs are essentially alike. As may be seen by reference to Fig. 2, the first three joints in *Anoplodactylus* are short and capable of comparatively little motion, while the fourth, fifth, and sixth joints are long, most of the movement of the leg taking place

¹ The second pair of appendages, the "palpi," are absent in *Anoplodactylus* and *Pallene*.

in these. The articulations are so arranged that there is little chance for motion outside the vertical plane, the leg thus moving up and down in the same plane in which it extends from the body. There is, however, a possibility of movement of the leg backward and forward in the horizontal plane to a certain extent, this motion occurring chiefly in the articulation between the first and second joints. The principal movement of the leg, that is to say the movement in the vertical plane, when the animal is swimming free, is shown approximately in the diagram. Starting with the leg in the position shown at *A*, and considering only that portion distal to the third joint, the next movement is essentially a straightening out and raising

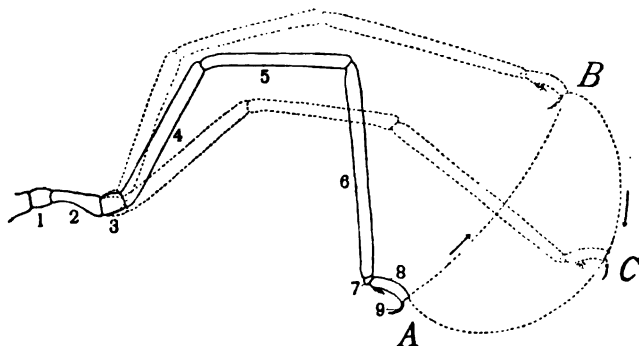


FIG. 2. — Diagram showing movement of leg in *Anoplodactylus*.

dorsally of that part, bringing it into a position as shown at *B*. The leg is now extended still farther and brought downward to *C*, then inward, and at the same time flexed, to the original position, *A*. This serves to indicate the movements in a rough way, and it can be readily seen that so long as the specimen is free in the water and all the eight legs are working with this same treading motion, the tendency is to propel the animal dorsalward, that is, in a direction perpendicular to its dorso-ventral plane; the fact that the legs extend radially from the body (Fig. 1) would help to keep the animal going straight in this direction, *provided they all beat with equal force*. If the claw of any leg should grasp a solid object as it comes down from *B* to *C*, the movement *C*–*A* would pull the animal in the direction of that object.

We are now in a position to examine more carefully the variations in the actions of the different individuals when placed in the water. As mentioned before, if the animal treads vigorously enough to overcome the force of gravity, it will swim; and so long as the body remains exactly in a horizontal position it will only move directly up or down according to the vigor of the strokes; but as soon as the body gets out of this plane the animal will progress through the water in the direction in which its dorsal surface is turned. For it to remain in a horizontal plane it is necessary that the legs should all beat with equal force; but as a matter of fact *the anterior legs beat oftener and with more vigor than the posterior legs*, thus raising the anterior end and tilting the animal backward. I did not make out any regular order of movement of the legs further than this, that the posterior legs seem to lack the vigor and strength and to be less under the control of the animal than the anterior pairs. A specimen which does not tread fast enough to raise itself from the bottom, or possibly one whose specific gravity is greater, crawls or walks straight ahead upon the sand, apparently, at first sight, much as an insect walks; but upon closer examination it may be seen that most of the movement is accomplished by the first pair of legs, assisted to some extent by the second, while the third and fourth pairs seem to be a hindrance rather than a help. By reference to Fig. 2 it can be seen how the anterior legs, by hooking into the sand, can pull the animal forward; while for the fourth pair to help in the forward movement it would be necessary for them to *push*, which would require a motion exactly the reverse of that which has been described for them when free from the bottom. Instead of this they drag along in a sort of helpless fashion, seeming to attempt the same movement as before, but hindered by striking the sand and by the forward movement of the animal as a whole due to the stronger anterior legs. A considerable backward and forward movement is now to be observed in the second and third legs, but this is probably also due to the pulling forward of the body after the legs are put down onto the sand, and not to a direct action of the legs themselves, the motion being allowed for, as before stated, by

the specially arranged articulation between the first and second joints. In order to prevent the specimens from swimming while making these observations, and to force them to crawl, a small collar of tinfoil was clasped around the body between the second and third pairs of legs, care being taken that it was small enough not to interfere with the movements.

Reaction to Light.

As has been stated, one of the most important factors concerning the movements of the Pycnogonids when placed in the water is the direction of the source of light. If the dish containing them is placed near a window, the animals either swim or crawl quickly to the light side of the dish.¹ This fact has been noted by Loeb,² who says of *Anoplodactylus*: "Es ist wie die meisten frei beweglichen Bewohner der Oberfläche des Meeres positiv heliotropisch," and he also states that when the body was severed between the second and third pairs of legs, the anterior portion still reacted in the same way, while the posterior portion, which was comparatively inactive, moved independently of the light. These results were easily verified, and it was further ascertained that the oculiferous tubercle (Figs. 1 and 4, *oc.*) is the photo-recipient organ, for when this was cut off the animals failed entirely to show any response to the light. Although there are, of course, individual variations, it is surprising how quick this response usually is, especially if the dish is covered above and on the sides away from the window, so as to exclude all light from other directions. In the diffuse light three or four feet from a northwest window, lively specimens usually traveled an average of 12 cm. to 15 cm. in thirty to forty seconds. The response seems to be more pronounced when the light enters horizontally.

In moving towards the light the animals may adopt any one of the modes of locomotion previously described: (1) they may

¹ This is not the case if there are hydroid stems or similar objects which the Pycnogonid can grasp. The tendency to cling to anything of this character seems to be stronger than the reaction to light.

² Loeb, J., "Bemerkungen über Regeneration," *Arch. Entwickl.-Mech.* Bd. ii, pp. 250-256. 1896-97.

swim entirely free ; (2) they may partially swim, kicking along on bottom with those legs that are down ; or (3) they may crawl with all the legs on the bottom. The second method is the most common and the one in which the greatest speed is made. But the striking thing to be noticed is that in the first and second methods, those in which they swim or partially swim, the movement is backwards, or nearly backwards, while in the third method of locomotion, when they crawl on the bottom, they invariably go straight ahead, that is to say, with the anterior end directed towards the source of light. It thus appears that in moving towards the light they orient themselves differently, according to whether they swim or crawl ; and, as I have shown, whether they swim or crawl depends directly upon the vigor or *rate* of the treading movement and not upon any difference in the *direction* of the stroke. The question naturally presents itself, Why should there be this difference in orientation in the two cases ? In order to determine this, let us first consider specimens which are forced to crawl by being weighted with tinfoil. If an animal so weighted is placed in the water with its anterior end towards the light, it crawls directly ahead without turning ; if its head is pointed in any other direction, it gains this same orientation by making a short circle, turning in the shortest direction towards the light. Now for any animal to walk in a circular path it is necessary for those legs on the outside of the circle to act with a greater force than those on the inside, and thus shove the body around ; in the case of an animal orienting itself so as to head towards the source of light, this means that *those legs away from the light act stronger than those towards the light*, they being the legs on the outside of the circle which the animal describes in coming around. If this rule holds true when the Pycnogonid swims, and I see no reason why it should not, we have a simple explanation of its orientation with reference to light at all times. This can perhaps best be made clear by taking a particular case and following it through. An animal is placed on the bottom with the long axis of the body at right angles to the rays of light. This is represented in the diagram *A* (Fig. 3), in which we are supposed to be looking

at one end of the Pycnogonid as it rests upon the sand. The arrow indicates the direction of the light. The animal at once begins to kick and the body is raised from the bottom, but since those legs on the side from the light (*a*) beat stronger than those towards the light (*b*), that side is raised more and the body is tilted so that the rays of light strike approximately perpendicular to the dorsal surface, as shown in *B*. Since the regular movement of the legs tends to propel the animal dorsalward, it moves toward the light. And now the fact that the anterior legs beat more efficiently than the posterior must be taken into account; this action tends to bring these legs uppermost, that is, around to the place of the legs (*a*) in diagram *B*. This is shown in *C*, where we see the animal from the side instead of endwise, as in *A* and *B*; in this position the posterior legs kick along on bottom. As a matter of fact the anterior legs seldom come entirely around so as to be directly uppermost, but only approximate that position, the third and fourth legs of one side or the other being the ones to touch bottom rather than the posterior legs; so that the animal does not move directly backwards but rather "corner-wise." If the movement is more vigorous, or if the light comes more from above, the animal may raise itself entirely free from the bottom, keeping, however, the same relative position.

If the orientation is different from the case given, it is easy to see how the same result is brought about. In a case where the animal is headed directly from the light, the anterior end has but to raise from the bottom to bring it into this position; and in the other possible case, when the head is directed towards the light, although the movements may be indefinite

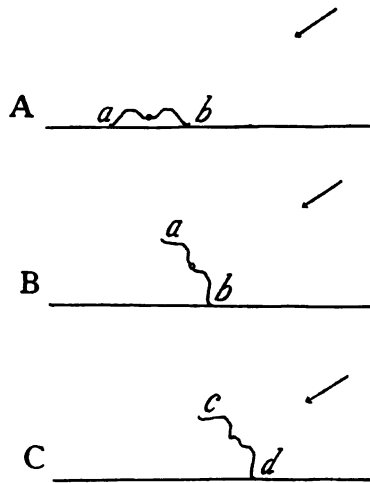


FIG. 3. — Diagram representing the reaction of *Anoplodactylus* to light.

at first, it soon gets out of direct orientation and then turns around the shortest way, as in the first case.

Pallene shows even a more marked positive phototaxis than *Anoplodactylus*. If the light comes in nearly horizontally, it usually tilts over to an angle of about ninety degrees, or until the body is nearly perpendicular to the bottom, and moves quickly towards the light by a rapid movement of the legs. Pallene is much the better swimmer of the two and seldom moves along the bottom in the manner described for *Anoplodactylus*. So far as I could make out, the leg movement is exactly similar in the two species.

Transfer of the Eggs.

During the latter half of August the males of *Anoplodactylus* may often be found bearing the egg-masses upon their ovigerous legs. As a general rule, among Pycnogonids the eggs are gathered into little spherical or spheroidal balls strung along on the ovigerous legs, but in *Anoplodactylus* they are in more or less irregular masses through which both of the ovig-

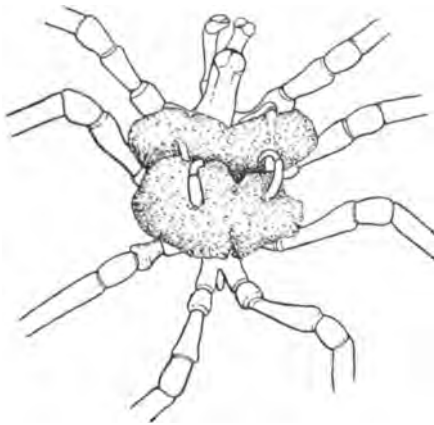


FIG. 4. — Male *A. lentus* from right side; walking legs removed. Reference letters as in Fig. 1.

crous legs pass (Fig. 4); their white color in this form, clearly offset by the dark body of the animal, gives them much the appearance of little bunches of wet cotton. Sars¹ says of the genus, evidently basing the statement on *A. petiolatus*, that there are "several globular egg-masses attached to the false legs in the male," and in his figure of this species

there are five such masses shown on the right ovigerous leg. It is possible that in *A. lentus* the irregular masses may later

¹ Sars, G. O., "Pycnogonidea," *The Norwegian North-Atlantic Expedition*, 1876-78. *Zoology*, vol. vi, p. 25, and Pl. II, Fig. 2 b. 1891.

roll up into separate balls, for I have had opportunity to observe them for only a few days after they were laid ; but this does not seem to me probable.

Cases of the males carrying the eggs are rare among animals and occur in widely separated groups ; the male of the obstetric toad (*Alytes obstetricans*) winds the egg-strings about his body and carries them till the tadpoles hatch ; the male of a South American frog (*Rhinoderma darwini*) takes the eggs into his vocal sacs to develop ; and the males of some of the Lophobranch fishes have brood pouches for the reception of the ova. In looking over the literature I have been unable to find any reference to this habit among the invertebrates, aside from the whole group of Pycnogonids. It seemed a matter of considerable interest to know just how such a seemingly intelligent act as the transfer of the eggs to the male takes place in animals whose movements in general seem to exhibit so low an order of psychic development, and I kept close watch of them with this point in view. So far as I am able to ascertain, the process has never been described, though Hoek¹ gives an account of the copulation in a European species as follows : "In regard to the way in which the eggs are laid, I had the good fortune to observe the copulation of a male and female *Phoxichilus laevis* Grube, when I was, last summer, in the zoölogical station of Professor H. de Lacaze-Duthiers at Roscoff. The eggs are fecundated the moment they are laid, and the copulation, therefore, is quite external, brought about by the genital openings of the two sexes being placed against each other. Half an hour after the beginning of copulation, the male had a large white egg-mass on one of his ovigerous legs, and about one hour later both masses were present." Only once, on August 16, at 6.15 A.M., was I fortunate enough to observe the pairing of *Anoplodactylus*. When first noticed both animals were among the hydroids ; the male was clinging to the dorsal surface of the female and headed in the same direction. Both animals were kicking slowly in an indefinite sort of way, but gradually the male drew forward and,

¹ Hoek, P. P. C., "Report on the Pycnogonida, dredged by H. M. S. *Challenger* during the years 1873-76," *Challenger Reports. Zoölogy*, vol. iii, p. 131.

passing down over the anterior end of the female, came to lie beneath her, the animals being now headed in opposite directions and with their ventral surfaces opposed. The basal joints of the legs of the female were approximated below, with the mass of eggs between them. As the male came around below the female, the ovigerous legs, which are curved at the ends, forming a sort of hook (Fig. 5), fastened into the egg-

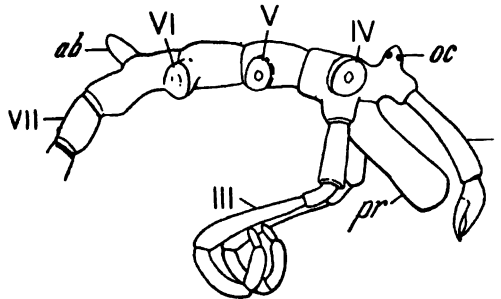


FIG. 5. — Male *A. lentus* from below, showing egg-masses on the ovigerous legs.

masses, and as the animals separated pulled the eggs away with them. The masses did not pull away clean, but strung out more or less, leaving a very few eggs still on the female. For some time after they separated

the male was observed to work the ovigerous legs slowly, the effect seeming to be to get the eggs more firmly upon them and into a more compact shape. The time from when the animals were first observed until they had separated was only about five minutes.

Some of the males have but one egg-mass on the ovigerous legs, but more often there are two, as shown in Fig. 4. I am unable to say with certainty whether this means that the male takes the eggs from two females or that he gets them in two masses from one; but from the fact that in those cases in which they were examined the eggs in the two masses appeared to be in the same stages of development, I am inclined to the latter view. The genital openings are situated on the ventral side of the second joint of all four pairs of legs, and it is easy to see how the eggs of one female might gather into more than one mass.

SUMMARY.

1. The three forms treated are more or less adapted in color to their several habitats.

2. Swimming is accomplished by a treading movement of the legs, which tends to propel the animal dorsward.

3. The stroke of each of the legs is the same in character, but is stronger in the anterior legs than in the posterior.

4. Crawling is accomplished by the same action of the legs as swimming, when the action is not strong enough to raise the animal from bottom. The anterior legs are most effective, pulling the animal forward; the action of the posterior legs is a hindrance.

5. Both *Anoplodactylus* and *Pallene* are strongly positively phototatic.

6. In *crawling* towards the light the animal proceeds with the anterior end in advance. If not oriented in this direction at first, it becomes so oriented by making a short circle, in every case towards the light. This means that those legs away from the light beat stronger than those towards the light.

7. In *swimming* towards the light the animal moves approximately backwards, with the anterior end somewhat raised. The amount it raises depends upon the activity of the individual and the slant of the rays of light.

8. This orientation is accomplished by the same actions that produce orientation when crawling, except that they are more vigorous, raising the animal from the bottom.

9. The transfer of the eggs from the female to the male is a comparatively simple process.

ANN ARBOR, MICH., Nov. 23, 1900.

EXPERIMENTS ON CUTTING OFF PARTS OF THE COTYLEDONS OF PEA AND NASTURTIUM SEEDS.

ABEGAIL C. DIMON.

THE experiments to be described were undertaken as bearing upon the general problem of the relation of food supply to growth. They were carried on under the guidance of Professor T. H. Morgan, to whom I am much indebted for aid and suggestions. The variations in food supply were produced in the pea and nasturtium, both dicotyledonous plants, by cutting off part of the cotyledons, thereby reducing the amount of food stored up by the parent plant for the use of the seedling. The questions that arise relate to the effect upon the size of the seedling, upon the differentiation of its organs, and upon the number and size of the component cells, caused by thus reducing the food supply. These questions may be answered from the results of the experiments.

As already stated, the pea and the nasturtium were the plants selected. Before deciding on them, however, the morning-glory, sweet-pea, radish, common bean, buckwheat, mustard, cucumber, and pumpkin were tested as to their suitability, by planting specimens of each with portions of their cotyledons cut off. It was found that the peas and nasturtiums, possessing large cotyledons, were more easily manipulated, and that their seedlings were hardier than those of the other plants. Their seedlings, moreover, grew rapidly, so that differences in the relative size of the plants were early noticeable and were well marked. Under favorable conditions, however, good results might be obtained from some of the other species, and it would be interesting to see to what extent they corroborate those from the two plants here discussed.

The seeds tested for availability, and subsequently all the pea and nasturtium seeds, were treated as follows : After they

had been soaked in water for from twelve to twenty-four hours to soften them, their seed-coats were removed and, from some of the seeds, parts of the cotyledons were cut off with a sharp scalpel, while the others were left in their normal condition save for the removal of their seed-coats. The normal seeds underwent the soaking in water and removal of the seed-coats to make their condition like that of the others except in the one point of food supply. When thus prepared, all these seeds were planted on sawdust, which was kept wet during their growth. In the case of the pea and nasturtium, as soon as the plants from these seeds were well started a second lot of normal seeds, treated in the same way as the normal seeds described above, was planted. In a short time, usually about two weeks, the plants of the second lot were found to be about the same size as those of the first lot that had come from the reduced seeds. Sections of the stems were then cut freehand, and camera drawings made.

The amount of cotyledon cut off from the seeds varied very considerably, and was not in all cases quantitatively determined. The variation may be seen from Table I, where are

TABLE I.

PART BY WEIGHT LEFT, EXPRESSED IN PERCENTAGES.	NO. OF PEAS.	NO. OF NAS- TURTIIUMS.	PART BY WEIGHT LEFT, EXPRESSED IN PERCENTAGES.	NO. OF PEAS.	NO. OF NAS- TURTIIUMS.
10-15	—	2	31-35	14	5
16-20	10	9	36-40	5	1
21-25	8	9	41-45	3	1
26-30	16	8	46-50	1	—

represented the percentages by weight of the part left in the case of 57 peas and 35 nasturtiums. The seeds were weighed after the removal of the seed-coats and again after the removal of part of the cotyledons, and the percentages express the ratio between the two weights. The percentages in the case of the peas vary from 16 to 48, with more than half the individuals between 26% and 35%; those in the case

of the nasturtiums vary from 10 to 41, with more than half the individuals between 16% and 25%, and nearly three-quarters between 16% and 30%. The chances, then, that from 65% to 74% of the bulk of any pea seed has been removed, are even; and the chances are three to one that from 70% to 84% of any nasturtium seed has been removed.

The variation in the amount of cotyledon removed appeared to influence the rate of growth of the seedling, but the number of plants of which a quantitative record of the development was kept was too small to justify an attempt to lay down a rule concerning the extent of this influence. The seedlings will therefore be regarded as belonging to only two classes, the normal and the dwarf, the latter composed of plants growing from seeds that have been reduced by removing part of their cotyledons. Plants of the two classes sprouted at about the same time, and for a short time the differences between them were not striking. As soon, however, as leaves began to develop, the normal seedlings shot ahead, surpassing the dwarf seedlings not only in size, but also in the number and size of their leaves. A comparison of two groups of pea plants, as given in Table II, from readings taken at three different times, shows the relative rate of development. The figures represent the height of the plants in millimeters and their number of leaves, while at the foot of each column is placed the average of the readings in that column. The readings from the dwarf seeds are arranged in the order of the fraction of the seed that is used for planting, and the readings from the normal seeds, in the order of the weight of the seeds, the smallest fraction and the smallest weight being at the top of their respective columns. The first period, two weeks after the seeds were planted, corresponds to an early stage of development; the second, five weeks old, is the stage just before the production of flowers by the normal plant; and the third period, nine weeks old, is the period of maturity, when the plants are bearing flowers and seeds. The letters *f.* and the word *pod* in the column marked "leaves," mean that the plant against which they are placed has a flower or a seed-pod.

TABLE II.

	TWO WEEKS.				FIVE WEEKS.				NINE WEEKS.			
	Dwarf.		Normal.		Dwarf.		Normal.		Dwarf.		Normal.	
	mm.	Leaves.	mm.	Leaves.	mm.	Leaves.	mm.	Leaves.	mm.		mm.	
a	17	5	35	11	22	9	77	23	35	—	95	fl.
b	10	—	29	11	29	9	70	19	47	—	89	—
c	16	5	32	13	35	15	86	27	44	—	108	pod.
d	22	7	32	11	26	9	77	27	35	—	102	fl.
e	26	11	32	11	54	20	89	27	70	fl.	117	pod.
f	26	9	38	13	47	17	77	27	63	fl.	95	—
g	29	9	41	13	60	21	92	28 fl.	77	pod.	108	pod.
Av.	22	7	34	12	39	14	81	25	53		102	

From Table II we may collect several facts as to the relations in size and development between dwarf and normal plants. In the first place, the ratio of height of dwarf to height of normal, expressed in percentages, varies as follows: At two weeks, 65%; at five weeks, 36%; at nine weeks, 52%. The ratios between the number of leaves of the dwarf and of the normal are 58% and 56% for the two and five weeks old plants, respectively. No readings were made of the number of leaves at the third stage, because some of the plants had lost leaves, and the oldest leaves of all the plants were beginning to wither, so that the number had not as much significance as in the earlier stages. Similar measurements of height and number of leaves made on another lot of peas showed similar variations in ratios, so we may consider the readings here given as typical. The advantage in size, then, which the normal seedlings show in the first stage becomes more marked as the plants grow older, while the differentiation, so far as expressed by the number of leaves, though markedly greater in the normal than in the dwarf plant, does not increase much faster in proportion in the former than in the latter. The time of the first appearance of flowers is, however, more significant of the differentiation of the plant than is the number of leaves, for the latter may depend on the size of the plant, as well as on its degree of development. The time of flowering was unmistakably earlier in the normal plants, for the first flower appeared upon them at five weeks, whereas the first flowers appeared on the dwarf plants at eight weeks. At this time three dwarf plants blossomed, while during the same period five of the normal plants had borne flowers, of which three had developed into pods. These facts suggest the conclusion that the normal plants not only are larger than are the dwarf plants but develop more quickly than do the latter.

It remains, therefore, to be seen whether the foregoing conclusions to which the macroscopic examination has led are supported by the examinations of sections under the microscope. An examination of this sort may be looked to to answer the following four questions:

A. What is the relative size of the cross-section of a normal and of a dwarf plant?

B. What is the relation between the number of cells in a normal cross-section and a dwarf one?

C. What is the relation between a normal and a dwarf plant, as regards the size of the cells?

D. How does the degree of differentiation of the normal compare with that of the dwarf plant?

Attempts were made to compare sections of the dwarf seedling under the microscope with sections of a normal seedling of the same size, as a check, as well as with the normal seedling of the same age, but the only specimens of check seedling available for comparison were so much larger than the dwarf that allowance must in every case be made for a discrepancy. The cross-sections studied were all cut free-hand from a level less than an inch from the ground in the growing plant.

The first question in the preceding paragraph relates to the relative size of the cross-section of the stem of the normal and dwarf plants. In the cross-sections represented by the figures the diameters of the peas have the relative values of 36 and 53, or the diameter of the dwarf pea is .68 as great as the diameter of the normal; the diameters of the nasturtiums have the relative values of 30 and 38, or the diameter of the dwarf is .79 as great as that of the normal. Cross-sections of the stem of the check plants have the values 46 and 33 for the diameters of the pea and nasturtium, respectively. The dwarf and normal plants were five weeks old and the check plant two and one-half weeks old. The plants selected for examination were typical ones, and the fact that the ratio between the diameters does not correspond to the average ratios between the heights given in Table II may be explained in two ways:

1. The ratio between the heights, as was seen, decreases with the increasing development of the plants, and the degree of development of the plants from which the cross-sections were taken was probably not the same as that of the plants measured in Table II.

2. The stem of small plants is always thicker in proportion to the size of the plant than that of large plants, so less difference in cross-section than in height is to be expected. Interpret this discrepancy as we may, the fact remains that the stem of a normal plant has a greater diameter than the stem of a plant sprung from a seed part of whose cotyledons has been cut off.

The ratio between the number of cells in a normal cross-section and a dwarf cross-section can be determined by counting the number in a definite sector of each. The results of counting the number of cells in a sector of 30° are : in the normal pea (Fig. 1), 410 cells ; in the dwarf pea (Fig. 2), 311 cells ; in the normal nasturtium (Fig. 3), 223 cells ; in the dwarf nasturtium (Fig. 4), 208 cells ; in the check pea, 404 cells ; in the check nasturtium, 219 cells. The dwarf plant has, therefore, decidedly fewer cells than the normal ; in the case of the pea .76 as many, and in the case of the nasturtium .93 as many. If the ratio between the number of cells was the same as the ratio between the diameters of the cross-sections, it would mean that the cells must be of the same size ; since the former ratio is larger for both pea and nasturtium, it means that the cells of the normal are larger than those of the dwarf plant.

The conclusion as to the size of cells in dwarf and normal plants may be confirmed directly by counting the number of cells in a definite area. Proceeding to do this for Parenchyma cells, the following results were obtained: In the normal pea (Fig. 1), 22 cells ; in the dwarf pea (Fig. 2), 43 cells ; in the normal nasturtium (Fig. 3), 21 cells ; in the dwarf nasturtium (Fig. 4), 32 cells ; in the check pea, 50 cells ; in the check nasturtium, 29 cells. The regions counted were all in the same part of the stem, and other counts were made that corroborate the figures here given. These figures confirm the conclusion reached in the preceding paragraph as to the size of the cells in normal and dwarf plants ; but in the case of the check pea plant it is found that the cells are smaller than those of the dwarf plant.

Though the statistics from microscopic examination that

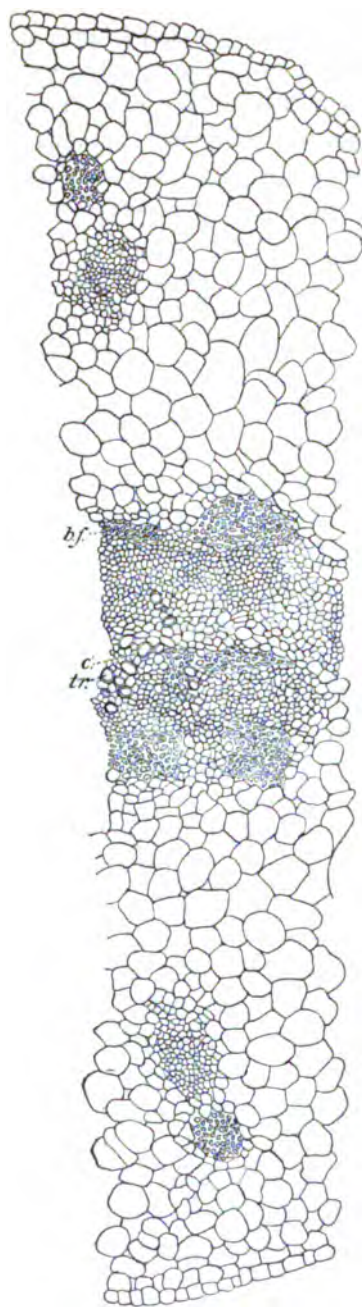


FIG. 1.

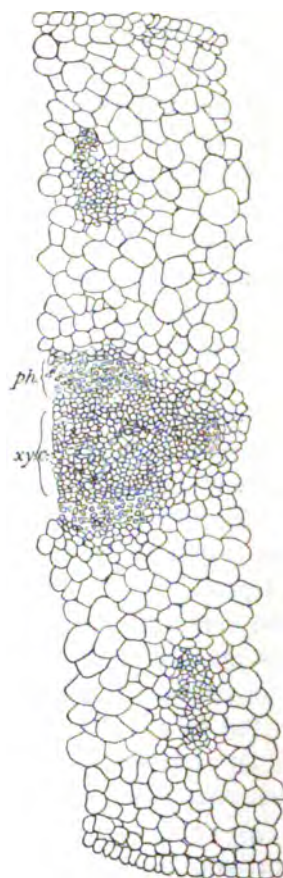


FIG. 2.

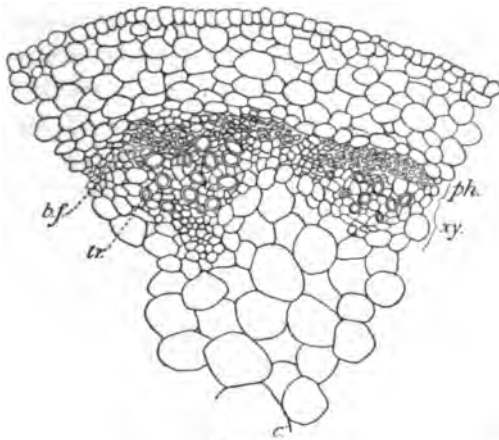


FIG. 3.

DESCRIPTION OF FIGURES.

b.f. = bast fibers. *tr.* = tracheid.
ph. = phloem. *xy.* = xylem.
c = center of stem.

FIG. 1. — Part of a cross-section of the stem of a normal pea seedling.

FIG. 2. — Part of a cross-section of the stem of a dwarf pea seedling.

FIG. 3. — Cross-section of a sector of the stem of a normal nasturtium seedling.

FIG. 4. — Part of a cross-section of the stem of a dwarf nasturtium seedling.

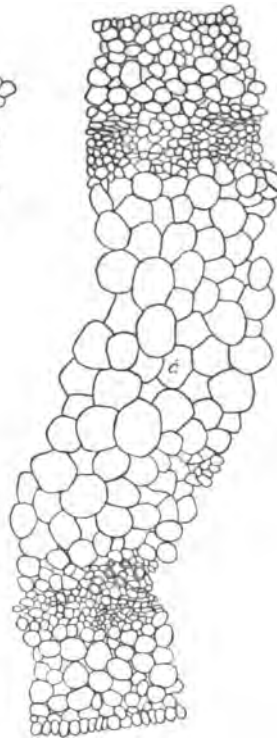


FIG. 4.

have been given and discussed were all drawn from three plants of each species, nevertheless that the general results are trustworthy is shown by another set of observations made on different pea plants. These gave the following results: Ratio of diameters of dwarf and normal plants, 111:62 (.56); cells in a given sector (7°), 165 in normal and 106 in dwarf (.64); cells in strips from center to circumference proportional in width to the size of the cross-section, 219 and 144 respectively (.66). The number of cells in a definite area was 29 in the normal and 44 in the dwarf. These measurements confirm those already discussed, for they show that the normal stem is larger in cross-section, is composed of a greater number of cells, and of larger cells than the dwarf.

The next question to be examined is the degree of differentiation of the various plants. This differentiation may be studied in the fibro-vascular bundles, where we may note the

appearance of the bundle as a whole, and the development of its elements. The appearance as a whole is indicated diagrammatically by the text-figures, which show especially marked differences in the case of the nasturtium. In the dwarf nasturtium the fibro-vascular elements are arranged in separate bundles around a central pith, while in the normal plant the phloem of the different bundles has run together, making a ring around the pith, the xylem being still discontinuous. Bast fibers seem highly developed with thick walls, and the tracheids are large, numerous, and clearly differentiated in the normal nasturtium stem; while in the dwarf stem the soft bast has just begun to show signs of thickening into fibers, and the tracheids are small, and comparatively few and poorly differentiated. In both specimens of pea the fibro-vascular

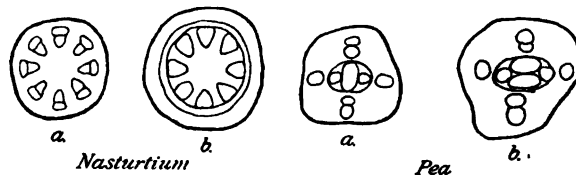


FIG. 5.

elements are arranged in an aggregate in the center of the stem and in four small groups peripheral to the large group. These bundles are more distinct and woody-looking to the naked eye in the normal than in the dwarf plants, and both bast fibers and tracheids are more numerous and highly differentiated. The normal plant, therefore, is more highly differentiated than the dwarf, as well as larger.

The conclusions reached from macroscopic and from microscopic examinations are then in accord with one another, and may be summarized by the statement that the removing of part of the cotyledons of a seed retards not merely the growth in size of the plant produced from that seed, but also its development. The plant, however, is not the counterpart of a younger normal plant, for it was found from comparing dwarf plants with check plants that the dwarf plant of a certain height was further developed than the check of the same height. The same point is illustrated by the fact that

a full-grown dwarf plant is smaller than a full-grown normal plant, as is shown by the nine-weeks stage of Table II.

The effect, it would seem, of removing a part of its food supply from the seed is not merely a transient one, but is one that can be traced through the whole life of the plant, and even increases as the plant grows older. The amount of food supply in the cotyledons influences, perhaps, the early stages of growth, while as the plant increases in size it becomes more and more vigorous and tends to grow more and more rapidly; so that a plant that is given an advantage over its fellow at the start will increase this advantage during subsequent development.

VARIATION AMONG HYDROMEDUSAE.

CHAS. W. HARGITT.

THE announcement of Bateson ('94), that "in the whole range of natural history there is no more striking case of the discontinuity and perfection of meristic variation than in the genus *Sarsia*, and the further proposition whether it is a mere coincidence that the specimens presenting this variation, so rare among the free-swimming Hydromedusae, should have been members of the same genus," directed my attention to this particular problem in conjunction with work upon this group of coelenterates which had engaged my attention for several years.

During the following years, therefore, collections of free medusae of several genera were made with a view to testing the problem raised by this observer. While as yet these collections are not extensive, except in a few genera, certain facts have been secured which may not be without value in their general bearing upon this as well as still broader problems of variation in general.

My collections have been restricted chiefly to the genera *Eucope*, *Obelia*, *Margelis*, *Pennaria*, *Gonionemus*, *Coryne* (*Sarsia*), and *Hybocodon*; the specimens of several others have been casually examined. Of the genera named, *Obelia* has not as yet been examined in sufficient numbers and detail to warrant any specific mention in this connection. And since these observations have been under way a paper by Agassiz and Woodworth ('96) on "Some Variations in the Genus *Eucope*" has appeared which so fully covers the facts involved in members of that genus, and are so coincident with my own, that no special details will be offered in connection with it, though the materials at hand are more abundant than upon any of the others.

What I shall have to offer in this paper, therefore, will be upon the other genera named, namely, *Coryne*, *Gonionemus*, *Hybocodon*, *Pennaria*, *Nemopsis*, and *Margelis*.

Coryne.

Of specimens of *Coryne* a comparatively few were available, though they were examined with unusual interest and care as belonging to the genus to which, apparently among the earliest, references to variation among Hydromedusae were made, and which called out the rather remarkable proposition of Bateson quoted in the opening paragraph of this paper. While the specimens were too few to warrant any definite conclusions, they nevertheless showed a most remarkable constancy in every morphological feature, not a single specimen exhibiting the slightest variation in any of the more conspicuous features, as tentacles, radial canals, manubrium, etc. If this constancy is as marked in different regions of distribution and for the large numbers cited by Bateson, it is not strange that he should refer to the matter in the terms quoted, as it would seem to be among the least variable of the free-swimming medusae of this group. It will at the same time show how very unsafe must be any such conclusion taken from so limited a range of observation.

Hybocodon.

Of the genus *Hybocodon* Ag. the number of specimens at my command has likewise been somewhat limited, slightly less than two hundred, still they have been sufficient to show some variation in certain features. This genus was instituted by L. Agassiz ('62), under which he included a Hydromedusa of very unique characters (*cf. Contribution to the Natural History of the United States*, Vol. IV, p. 243), one of which is the proliferous budding of medusae from the hydranth, which in turn give rise to secondary and many later specimens by a similar process of budding. (*Cf. op. cit.*, Pl. XXV, Fig. 13.)

The specimens which came into my hands were all preserved in formalin and had in consequence suffered considerable

distortion in the process, whereby minute variations of organs often became difficult of detection, yet I was able to demonstrate a fair degree of constancy in the general form of the medusa, its radial canals, tentacles, etc. I desire to direct attention to the number of tentacles. As stated by Agassiz, there seems to be a single long tentacle arising from the margin of the bell at the terminus of one of the radial canals, from the base of which arose later the proliferous medusae-buds, as shown in the figure already cited. From these secondary medusae other tentacles arose, giving to them the exact morphological equivalent of the primary or mother medusa. Hence, as several of these proliferous specimens budded off from the base of the primary tentacle, several tentacles would come to be clustered near the same point, giving the impression of a bunch of tentacles of the same nature. In several specimens in which the medusae-buds had not yet appeared, or could be detected as mere papilla-like bodies, these secondary tentacles were nevertheless well developed, and of a length frequently equal to that of the primary one. Now whether this be a variation, or whether it may not be rather fundamental, arising as a source from which the medusae are to spring, may perhaps be an open question, to be settled by a more critical examination of their development. Proceeding on the assumption before stated, I venture to cite it as a case of variation, though it may later be found to be rather the normal process.

The rather unusual character of this medusa, both in its origin and proliferous progeny, led me to suspect that it might exhibit more than the usual phases of variation; but in this I have been disappointed, except in the point just cited, — its constancy in almost every morphological detail being quite marked. As stated, however, in connection with observations upon *Coryne*, the limited number of specimens examined, and furthermore their distortion due to preservation, are barriers which should suggest reasonable caution in the formulation of any conclusion.

Pennaria. — Of this medusa I have had an almost unlimited number of specimens, having collected them during three

years devoted to the embryology of the species common in the waters of the Massachusetts coast,—*P. Tiarella* McCr. A critical study of these medusae is, however, rendered difficult and tedious owing to their minuteness and form. In size they are only about .8 mm. in diameter by about 1.5 mm. in length. The highly oval form renders difficult a study of the aboral surface and the junction of the radial, or chymiferous canals,—a point of considerable variability in many cases in other genera, notably *Gonionemus*, to be noted later. In a study of their morphology Smallwood ('99) has pointed out the variability in the structure and development of these canals. He has shown that in a considerable proportion of specimens there is a tendency to atrophy both in the radial and circumferential canals, especially the latter. These changes are not evident in a surface study of specimens, the pigmentation which marks their course being fairly constant. The principal variation to which I desire to direct attention in this connection is a physiological one, *viz.*, a rather marked variation in habit and activity. I have discussed elsewhere this feature ('00) and need only refer here in brief to those observations. As there pointed out, there seem to be two rather distinct features of habit; namely, a rather deep-water habit upon rocks, seaweed, piles, etc., and a surface habit upon eel-grass or similar support, which serves to bring the colonies to the surface, thus often in a low tide exposing them directly to the action of the midsummer sun and temperature.

Associated with these differences are correlated variations in the form and color of the colonies, or, as Bateson would designate them, "substantive variation." The surface or eel-grass varieties exhibit more distinctly the pinnatifid character which marks its specific peculiarity, due doubtless, in part at least, to the prone or floating disposition of the colonies. Associated also with this is the much higher coloration so conspicuous in these specimens, a variation extending not only to the perisarc of the colonies but also to the medusae and the eggs, which are rather bright orange, while those taken from the deeper waters are a pale, creamy white, with the slightest trace of pink in many cases.

Of the further physiological differences one of the most marked is that of the relative activities of the medusae of the afore-mentioned varieties; those of the surface habit exhibiting a much greater degree of activity and other vital phenomena. These, as previously pointed out, are extremely active, being liberated from the hydranths promptly upon maturity, swimming with great ease and freedom, and discharging the sexual products with great promptness. On the other hand those of the deep-water habit are passive, or even sluggish, — in many cases the medusae never becoming free from the hydranth, — discharging the sexual products with much less regularity and ease, and dying very soon after. These medusae are short lived at best and never increase in size after liberation from the hydroid. I would suggest the probable correlation of some of these features of variation with the degenerative tendency shown in both structural and physiological variations already noted, especially in the atrophy of the chymiferous canals.

A histological study of the tentaculocysts likewise shows degenerative tendencies, as does also the very rudimentary condition of the tentacles, which are barely distinguished as bud-like protuberances upon the margins of the bell.

In connection with previous work upon the development of *Pennaria*, attention was directed to variation in the rate of cleavage and subsequent development. This would seem to be a matter of considerable interest in connection with the fundamental problems of physiological variation. It is well known, of course, that cleavage is a phenomenon subject to considerable variation as to rate, due to variable conditions, and to some extent independent of sensible differences of environment. It seems to me, however, that in the case of *Pennaria* there are presented such marked extremes in this respect that it may well be considered as in some measure correlated with other features of physiological variation.

The variable rate in the later phases of larval development is also worthy of note. From data obtained during three summers of observation the range of time involved in the larval history varies from about two days up to about two weeks.

While in most cases these observations were made upon specimens under artificial conditions, namely, aquaria of variable sizes, etc., still the variations occurring were exhibited by larvae under identical conditions, such as they were.

Under the head of abnormalities in immediate connection with these observations, attention was also directed to certain variations in the morphology of the larvae and early polyps. Among these may be mentioned

1. Twin-planulae, — planulae with bifurcated ends, irregular bud-like outgrowths, etc. (*Cf. op. cit.*, Figs. 4–6, and 8, Pl. 1.) It was suggested that they were probably due “to the intrinsic prepotency of hydroids to bud and branch.” While this is probably an explanation of the facts, that they exhibit interesting variations from the ordinary is not discredited on that account.

2. Attention was also directed to an interesting polyp form (*op. cit.*, Fig. 1 of text), which presented so marked a variation as to give rise to some doubt concerning its Pennarian affinities. In view of the rather large range of variability exhibited by the medusoid and larval persons already considered, I am still convinced that this is only a further illustration of the same principle. Indeed, I have during the present summer observed in other polyps reared under similar conditions the same variation from total annulations to less and less degrees. A few additional annulations of the hydroid perisarc is matter of no special surprise. A complete annulation of the early and plastic colony, while quite unusual, need not be regarded as improbable or especially strange.

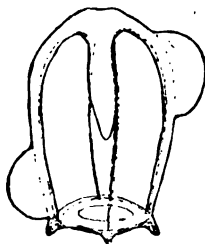


FIG. 1.

Another feature which may perhaps rather be designated as a monstrosity, or incidental excrescence, may be noted in this connection; namely, certain wartlike or pustular vesicles which often appear at various points on the exumbrella of the medusae. These are fairly represented in Fig. 1. The figure indicates relative positions where they are most likely to occur, though in no case have I noted more than one upon a given specimen. . . . A similar structure is referred to by Agassiz ('65) and

explained as due to distortion caused by the pressure of the ova within the bell. This I am convinced is a mistaken view, for I have noted it upon specimens both living and on those killed and preserved in formalin, in specimens with and without eggs. It seems, moreover, to be wholly restricted to the outer ectoderm only, in no case involving the inner ectoderm of the subumbrella. There is nothing indicative of the cause or character of these excrescences. Whether they are permanent or merely transient features I am not able to say, the short-lived condition rendering any determination difficult if not impossible.

Nemopsis.

Through the courtesy of Mr. Strong I had the privilege of examining a small collection of about one hundred specimens of *Nemopsis Bachei* taken in the tow off Tarpaulin Cove. The variations here seemed quite as evident as in *Eucope* and *Gonionemus*. Here again the variable features included radial canals, manubrium, gonads, and tentacles. Fully five per cent showed some feature of variation. About two per cent had but three radials and three gonads. One of these showed a definite correlation, including all the organs named above. One, however, of the trimerous forms had a fourth sensory bulb and tentacles, though these were less prominent than were the other three sets. The oral tentacles likewise shared in the correlation.

One specimen was a symmetrically pentamerous form with a perfect correlation of all the organs under consideration. Another specimen was quite as symmetrically hexamerous.

Several other specimens exhibited apparent symmetry in the number of gonads. Frequently one of the series showed the gonad of one canal very unequally developed as compared with the others. But while approaching sexual maturity, and in many cases fully so, it is of course impossible to say with certainty that the short gonad might not have shown further development with age. In any case it certainly showed variation as to *development*.

Concerning variation in the number and order of tentacles it is difficult to determine definitely, since in *Nemopsis* they constantly increase in number as the medusa grows, much as in *Margelis*. So while there appears to be considerable variation in the number and arrangement it may be rather due to variable development than to any actual meristic variation. The same may be true as to the order of appearance. The paired, capitate tentacles at the apex of the bulb appear uniformly first and seem to be fairly constant. The latter filamentous tentacles appear to arise in pairs successively toward the margins of the bulb. Since I was not able to follow this development in the stages of growth of the medusa it is impossible to determine definitely this point. So while there is upon the preserved specimens considerable want of symmetry in this respect, yet it may be due in part to slightly variable rates of development. No constancy was apparent in the matter, and it would seem therefore to be physiological rather than morphological.

Similarly there was apparently some variation as to the number and distinctness of the otocysts upon the sensory bulbs. Normally there is a single eye-spot at the base of each tentacle. But in many cases they were apparently absent. And while it is not impossible that they had been rendered indistinct by the formalin in which they were preserved, still it remains quite certain that marked differences were distinguishable among various specimens of similar size and preservation, and perhaps only critical histological examination will be adequate to finally determine this point, and this I have not been able to make.

Margelis.

Of these medusae more than five hundred specimens have been examined, most of which were quite young, having only the four pairs of marginal tentacles and four unbranched oral tentacles and measuring only about 0.5 mm. in diameter. Typically this medusa may be characterized as having a high hemispherical bell, four radial canals, at the distal or marginal ends of which four clusters of filiform tentacles arise. The bell is

thick, velum not specially prominent. Manubrium subconical, bearing four oral tentacles which divide dichotomously into small clusters of tentacles. Figs. 2-4 give good general impressions of the animal. The very minute size made necessary the constant use of the compound microscope in all the examinations.

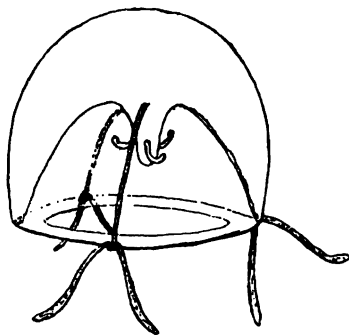


FIG. 2.

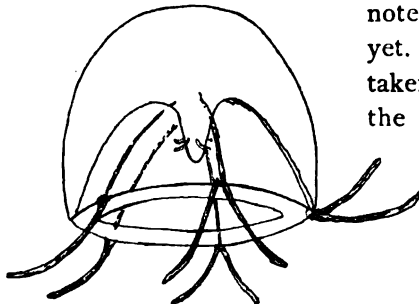


FIG. 3.

The variation in radial canals was usually correlated with that of the tentacles. In Fig. 2 is shown a trimerous specimen, in which there was a perfect correlation of all the organs, including oral tentacles (several specimens noted). In Figs. 3 and 4 is shown a pentamerous form, in which there appeared little meristic correlation.

For example, it will be seen that while five sets of tentacles correspond with the five radial canals, two had but a

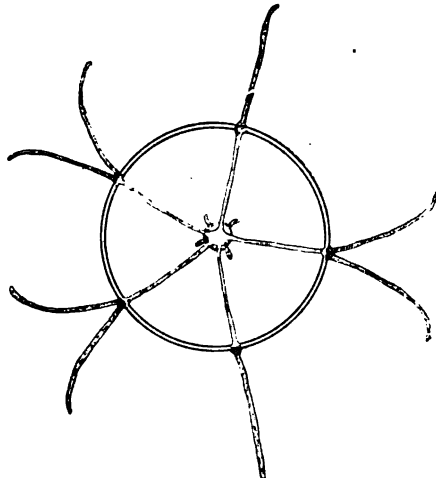


FIG. 4.

single tentacle each, while the other three sets contained the normal (at this stage) number, two. A point not shown definitely in the figure is the fact of only four oral tentacles.

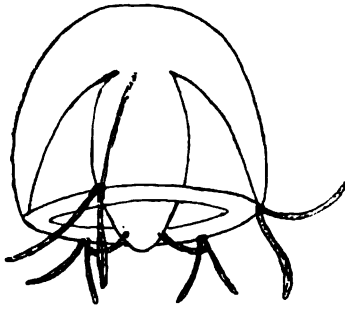


FIG. 5.

In Fig. 5 is shown a specimen with monstrously developed manubrium, protruding beyond the velum, and provided with but two tentacles. In several specimens similar enlargements were noted, though not so pronounced as in this, except in certain cases, apparently pathological, in which the entire bell was evaginated and greatly shrunken, with the manu-

brium greatly enlarged. No account was taken of such specimens, for they were evidently due to conditions other than those of health. It may be noted in this connection that several specimens were found exhibiting similar vesicular or pustular enlargement to those observed in connection with accounts of *Pennaria*. Here is further evidence, if such were necessary, that these enlargements, resembling distortions, could not have been produced by the enlarged gonads, for in the species under consideration the sexual organs were as yet undeveloped.

In Fig. 6 is shown in diagram an aboral view of a condition found in several specimens, in which there seem to be secondary, peripherally directed radial canals, extending nearly half-way over the bell. As this medusa has normally but four such canals this is a well-marked case of variation in the direction of a condition quite common in *Rhegmatores* and many trachomedusae. I regret that none of the specimens to which I have had access are of approximate maturity so that such incipient variations might be traced forward to their perfection, in order to ascertain

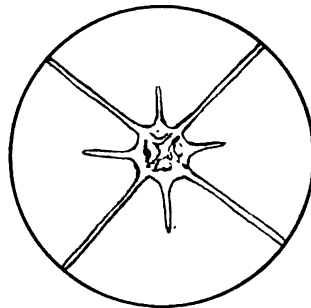


FIG. 6.

whether additional sets of tentacles, etc., would be found correlated therewith.

The origin and symmetrical interpolation of such secondary canals is very different, it seems to me, from that arising from the bifurcation seen in *Eucope* and *Gonionemus*, though their function in the economy of the organism may be the same.

Only one phenomenon more will be discussed in this connection, namely, that of double or twin medusae, which is shown in Fig. 7. Only one specimen of this character was found in the entire lot. In every respect — size, general form, organic relations, etc., the double feature alone excepted — the specimen seemed fairly normal, having this one further feature, that one was a trimerous specimen and that one set of tentacles contained three as against the

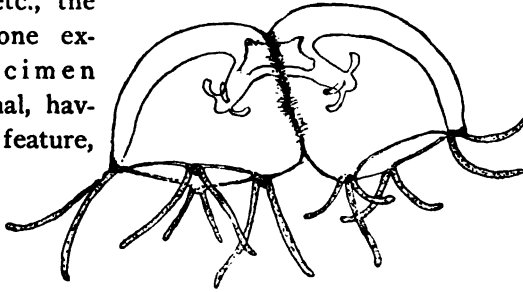


FIG. 7.

two each of the other sets. The furrow-like depression along the line of union presents the aspects of late coalescence similar to that involved in artificial grafting (*cf. Biological Bulletin*, Vol. I, p. 41).

But a more critical examination shows that the union is much more profound, involving the gastric cavity and therefore the whole chymiferous system.

The specimen was not seen alive, and hence nothing can be said as to coördination of physiological activities, mode of progression, etc. But from what has been proved from experimentation on these points (*cf. op. cit.*) it may safely be inferred that similar coördination at least prevailed in such a case as that under consideration.

Podocoryne.

Of medusae of this genus I have had at my command only a few more than one hundred specimens, a number too small

to warrant any formal deductions, but taken in connection with others it will not be amiss as showing the facts of variation pertaining to this genus.

Like Margelis this medusa is quite small when liberated, indeed never attains a size of more than 1 mm. in diameter, so far as I am aware. In general, it is similar in organization to Margelis, has four radial canals and four primary tentacles, between which a second series of the same number soon arises. Like Margelis there is apparently a fair degree of constancy in meristic features. Three trimerous specimens occurred in the lot, which comprised the extent of variations along this line. There were, however, considerable variations of size, form, etc., which increased the total to at least five per cent. It must not be forgotten, however, that the extreme minuteness of specimens, necessitating the constant use of the microscope, might easily involve an oversight of specimens in sufficient numbers to materially raise this ratio of variation.

Gonionemus.

It is among the members of this genus that I have been able to work out the most numerous and detailed series of variations. The total number of specimens of *Gonionemus* examined during the progress of the work was more than two thousand. (Of the first series studied no record was kept.) While smaller than the number of *Eucope*, to which reference has been made, the number is yet sufficiently large to insure against an unusual per cent which might be due to local or other incidental influences. Moreover, the collections were made during three summers, and by several collectors, so that the results obtained may be considered as closely approximating the actual state of variation now under way. It is a pleasure to acknowledge in this connection courtesies from Messrs. Coe, Parker, Perkins, Gray, and others for permission to examine collections of these medusae made by them, which has facilitated my work.

In these studies attention has been directed chiefly to the following structures : 1, radial canals ; 2, gonads ; 3, manubrium ;

4, tentacles; 5, otocysts. Other features of subsidiary nature will have mention in their appropriate places.

It ought to be stated in passing to details and tabulation of results that only in the matter of radial canals and gonads have the entire two thousand specimens been examined, and not all of those with equal detail in each case. In number and variation of tentacles, spurs, anastomosing of canals, etc., the tabulations were limited to one thousand; and in the case of otocysts to less than one hundred, a reason for which will be given in the appropriate place. I may also state that owing to the insignificant sexual differences, usually requiring microscopic examination to certainly decide, no effort has been made to determine the relation of variation to sex.

Gonionemus is a rather well-defined trachomedusa, first definitely described from the Atlantic coast by Murbach ('95). It is characterized by the typical four radial canals, which have the folded gonads suspended along their under surfaces, cruciform manubrium, as seen in transverse section, normally with four oral lobes and sinuously folded lips. Tentacles are numerous and similar, each characterized by a suctorial bulb near the tip, beyond which it often makes a sharp bend.

These medusae abound from June to October in a small pond near the Marine Laboratory, known as the "eel pond," which communicates with the open harbor by a narrow inlet. Lately they have been taken in the outer harbor, though in small numbers. I may mention this fact of the localized habitat, since it may well be a question whether its peculiarity may not be an important factor in the physiological aspects of the variations to be considered.

Taking up now the consideration of the several points in the order given, attention will first be directed to the

Radial Canals.

On this point an estimation of the ratio of numerical variation based upon fifteen hundred specimens gave nearly five per cent (4.82). On the matter of their form or disposition, *i.e.*, whether in their course to the marginal canal the bells

were divided into symmetrical segments, the results showed no less than thirty per cent of variation. On the variations shown in their aboral confluence, or union with the gastric pouch, the calculations gave 14.4 per cent.

Of the entire number examined only a single specimen was found having but two radial canals and two gonads. These were at an angle of 180 degrees, *i.e.*, dividing the bell into two symmetrical halves, as shown in Fig. 8, excepting along the rela-

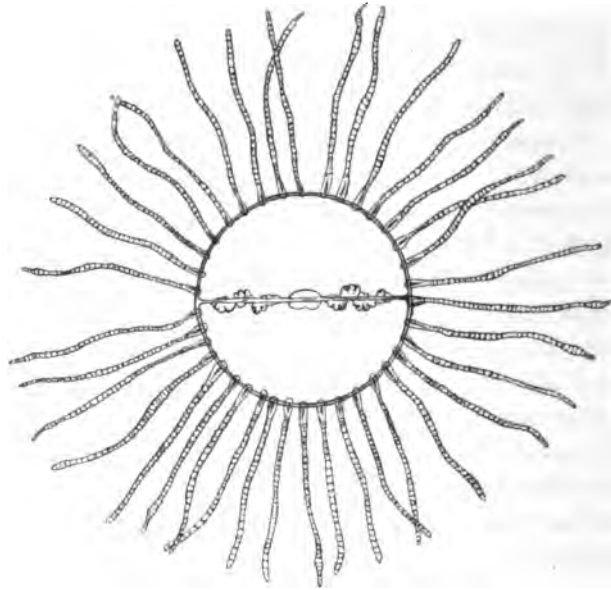


FIG. 8.

tive number of tentacles upon each half. One other specimen, however, was found having only two canals, similarly disposed and with a similar number and disposition of gonads; but in it there was a rather evident rudiment of a third springing from the peripheral end of one of the canals, thus destroying the bilateralism characteristic of the first.

The largest number of canals found was six. This, while much more common than those with two, was much less so than specimens with three and five. Seven specimens in all were found having this number, and in one of these the sixth was due to the evident forking of one of the five apparently

primary canals, which divided the bell into approximately pentamerous segments, as shown in Pl. III, Fig. 12. In another there was a very evident forking of two of the four primary canals, as shown in Pl. III, Fig. 9; for while the bell was divided into hexamerous segments, the manubrium was symmetrically tetramerous. In every one of the other five hexamerous specimens the canals converged at the aboral pole in a perfectly symmetrical way, though the hexamerism extended to the manubrium in only two specimens, and in these only

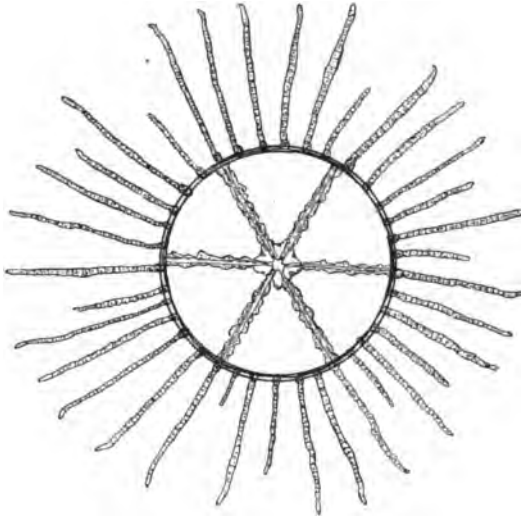


FIG. 9. — Hexamerous specimen showing pentamerous stomach and varying size and distribution of tentacles.

the basal portion or gastric pouch was strictly hexamerous. In at least two other of these hexamerous specimens the oral lobes of the manubrium were four (*cf.* Fig. 9).

Of specimens having three and five canals there were by far the larger number, with the preponderance slightly in favor of the trimerous variety, but not sufficiently so to warrant any conclusions as to the question whether the course of variation was toward a trimerous rather than a pentamerous condition. Of those with three canals there were twenty-one specimens, while of those with five there were eighteen specimens. Of those making up the total of seventy-two specimens there were

twenty-five specimens from distinctly tetramerous forms, having short forks less than one-third the length of the entire canal, or of such other variable aspects as to warrant their inclusion under this head. Pl. II, Figs. 6-9, and Pl. III, Figs. 1-5, will best illustrate this point.

It now remains to consider somewhat more in detail the individual variations exhibited by these several types. Directing attention first to those illustrated by the figures just cited, it may be seen that the variation here seems to be in several

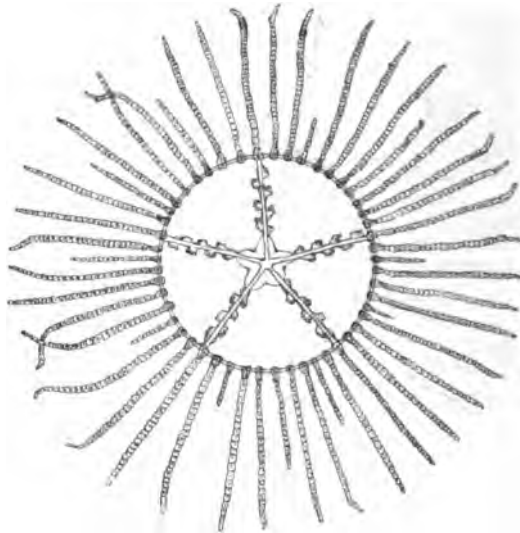


FIG. 10. — Symmetrically pentamerous specimen, but with the several series of tentacles appearing at irregular intervals.

directions. (1) Atrophy, as shown in Pl. II, Figs. 4, 6, 8, and 9. In all these the evidences of degeneration are quite clear. First, in Figs. 6 and 9 there is the atrophy of the connection of one of the canals with the gastric pouch and the correlated reduction of the fourth gastric pouch and the further failure of the obsolescent canal to develop its usual gonad, the merest rudiments of which are apparent. Furthermore, in the same figure there is shown a still further atrophy of a second canal, which extends only about halfway to the margin, and correlated with that fact is the associated imperfect development of its gonad. A still further illustration of this degenerative

tendency is shown in Pl. III, Fig. 6, where only the vestige of the fourth canal is shown, the reduction in extent of two others, with the further correlation of the evidently bilobed condition of the gastric pouch.

(2) *Asymmetry.* This is more or less consequent upon the atrophy already noted, as will be seen from a comparison of the figures just cited, and involves to a certain degree the entire organism, gastric and oral symmetry, no less than that of the

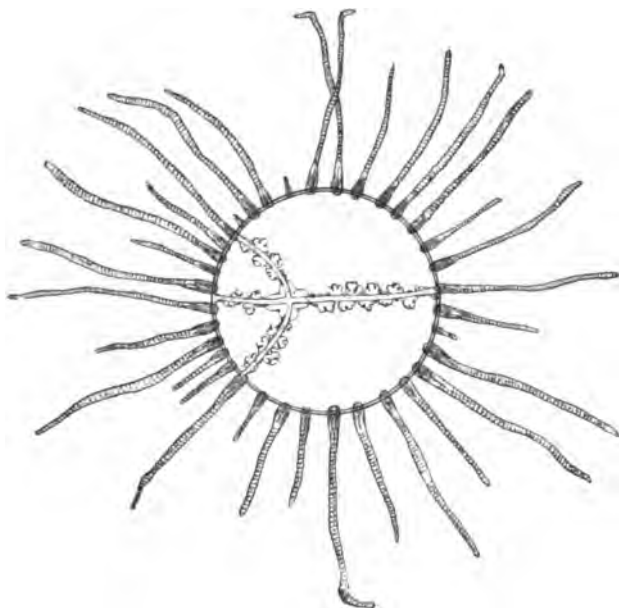


FIG. 11. — Tetramerous specimen of very unsymmetrical type.

bell and tentacles. Some further reflections on these lines will more naturally come up in connection with later discussions.

Another type of variation is shown in Pl. III, Figs. 1-5. In these specimens, while the tetramerous type is more or less evident from the number of canals, gonads, or gastric pouches, still there is a rather definite tendency toward a trimerous aspect of the medusa as a whole, so far as the segmentation of the body is concerned. In Fig. 1, while there is a clearly tetramerous condition exhibited which extends to the several organs involved, there is yet such an approximation of those

marked *a* and *b* as to leave the bell and number of tentacles in a closely trimerous symmetry. In Figs. 3-5 this evolution of trimerism is so evident that it would seem to point toward a preponderance of variation in this direction. As, however, will be seen later, facts of a very different kind seem to point as clearly in the opposite direction. It may as well be pointed out in this connection that the loopings of canals shown in the figures under consideration are variously simulated by

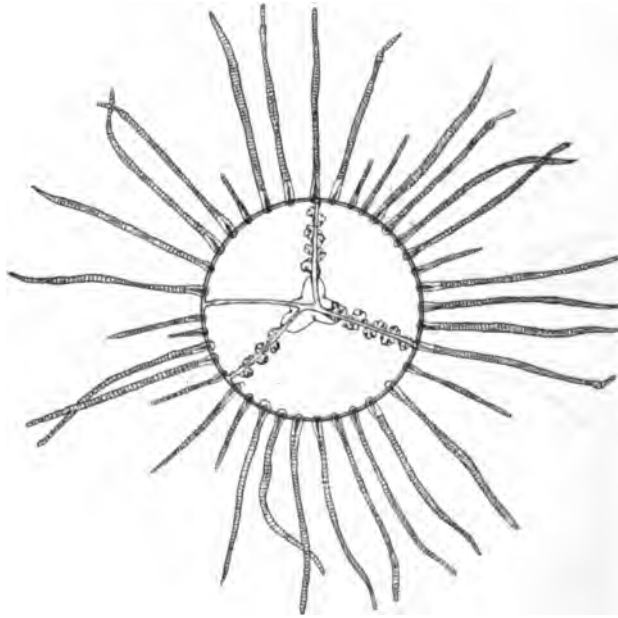


FIG. 12. — Specimen of very unsymmetrical character.

structures shown in Pl. II, Figs. 1-4, and 10. A critical comparison however, while showing many unusual features in these latter structures, will probably demonstrate their fundamental likeness; but this will be considered later.

We may next consider a type of variation fairly illustrated by Pl. III, Figs. 9 and 11. As will be seen from a glance, there is here exhibited a clearly defined tendency toward an increase in the number of canals, hardly less marked than that of decrease just considered. Indeed, specimens with bifurcated canals of this character were rather more common than those

of the last type, a fact, when taken in connection with the closely similar number of distinctly pentamerous forms, of great importance as showing that in neither case are we warranted in concluding that the course of variation is in one direction rather than in another. In order to reach anything approximating conclusiveness on this point a larger number of specimens studied through a successive series of years would be necessary. As I have already intimated, the collections forming the basis of the present discussion of this genus were made during at least four years, and while I have not made this a matter of critical comparison, it has not been at all apparent that during this period there has been any appreciable ratio of difference.

Passing now to the consideration of other aspects of variation evident in the canals, attention is next directed to their morphology. As is well known, the chymiferous canals in medusae are tubular structures of fairly constant size in members of the same species and of similar sizes, and their courses are usually direct from the center to the margin in most of the Hydromedusae. As Agassiz and Woodworth ('96) have shown in the case of *Eucope*, however, there are not a few departures from this rule. The same is true of *Gonionemus*, as a glance at Pls. I and II will demonstrate. Not only does the diameter of the canal vary greatly in many specimens, which is of only incidental concern, but in many cases, as in Pl. I, Figs. 9, 11, and 12, various loops and diverticula in the form of spurs are formed at various points and at various angles along their course. These are of varying sizes, lengths, etc., and were found on between one and two per cent of all the specimens examined. In the paper just cited the authors suggest in these facts a possible simulation of a condition "characteristic of the Discophores" (p. 122). Whether there is in these structures anything more than simulation or parallelism as compared with the Scyphomedusae, I shall not at present discuss.¹ As compared with the typical canal, however,

¹ It may not be amiss, however, to state in this connection that in *Rhegmatores* there is a much more evident correspondence or resemblance in this matter than in either *Gonionemus* or *Eucope*. While possessing a large number of

there seems to me to be little doubt that they are fundamentally of similar origin and function. While in many cases there are extremely small serrations of the canal walls, in other cases (Pl. I, Figs. 1 and 2; Pl. III, Figs. 4 and 5) they are more prominent, even occasionally forming anastomosing connections between adjacent canals. Similarly the loops already referred to are probably in most cases anastomosed spurs.

An examination of the several figures of Pls. I-III will bring to the attention some interesting and rather anomalous illustrations of another phase of the structures under consideration. As will be seen, there is here almost every degree of intergradation between the perfectly symmetrical cruciform aboral junction of the chymiferous canals and the perfectly circular canal about the base of the gastric pouch into which the radials connect before their connection with the gastric cavity. By careful injections through the radial canals I have clearly demonstrated a direct continuity of the chymiferous system throughout these several channels. Little doubt can therefore remain concerning the fundamentally similar character of these various structures. Nor is it more doubtful that in function they are fundamentally similar; and while concerning the question of their significance in relation to the affinities of the Hydro- and Scyphomedusae there may be room for wide difference of view, that they serve similar functions in both is highly probable, if not quite certain.

In passing to the consideration of a specimen of unusual form, it should be noted that in the origin of spurs, extra canals, etc., they were with very slight exceptions, which seem to me easily explained, centrifugal, *i.e.*, from the central toward the peripheral portions of the body. The apparent exceptions are shown in Pl. II, Figs. 4 and 7, where portions of canals extend from the margin toward the center. As will be noted, however, there are in both cases spurs from the central region in the line of the peripheral branches which would strongly

radial canals, many of them show bifurcations toward the margin, and in not a few cases are there found centripetally developing canals similar to those of *Car- marina*. This medusa likewise shows many other phases of variation, spurs, anastomoses, etc., of canals, but no details will be undertaken in this connection.

suggest that there had been complete unions of these partial canals at an earlier stage and that the present condition was the result of atrophy such as is shown in Figs. 6 and 9. It would seem therefore quite just to conclude that these several structures, spurs, partial canals, loops, etc., have had their development usually from the central pouches or canals and not from the peripheral or marginal canal.

The unusual specimen, to which reference is made in this connection, is shown in Fig. 13 of the text. It would appear to partake somewhat of the nature of a monstrosity and in some respects of the nature of a marginal bud, suggestive of a secondary medusa. Aside from the general form there is little to confirm this possibility; there is no sign of manubrium; and the canals and tentacles are quite continuous with those of the primary medusa. As will be noted, there are vestiges of gonads upon the peripheral termination of the median canal, while the branches are wholly devoid of any signs of such structures. Only a single specimen of this character was found and it exhibits another aspect of erratic variation.

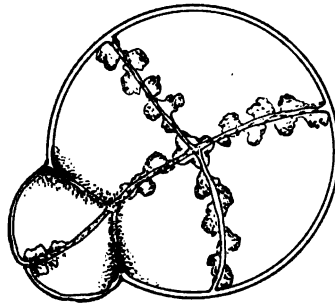


FIG. 13.

Gonads.

In the comparisons of gonads only specimens apparently sexually mature were taken (as noted before, no distinction was made between sexes). In the cases wholly devoid of gonads the size and other organic conditions were considered as sufficient to warrant the conclusion that they were probably of such age and general development as are usually correlated with perfect sexual maturity. In the whole number of specimens examined 3.6 per cent showed numerical variation of the gonads; of specimens with less than the normal number, two per cent; of those with more than the normal, 1.2 per cent; of specimens without trace of gonads, .4 per cent. As will be

seen in comparing these ratios with those concerning the radial canals, there is here again a slight tendency toward the smaller or trimerous forms, though not specially marked, especially when account is taken of the fact that only on specimens with more than the normal canals would additional gonads be found, while it was not rare to find in tetramerous forms specimens with only three gonads, or even less, one tetramerous specimen having a single gonad. Concerning variations of the several gonads of individual specimens no account was taken, owing to the difficulty of determining relative differences in organs loosely suspended in sinuous folds, as are these in *Goniomemus*, and by the further fact of the continued growth and successive discharge of the sexual products, as seems to be the case here.

Manubrium.

As in most medusae, the manubrium is a rather prominent and important organ. In correlation with the tetramerous organization of the medusa, the manubrium, including in this general term the basal gastric pouch and oral opening and lobes, is of similar form and adjustment. As will be noted, however, by a comparison of the several tables, there are many exceptions, or, in other words, considerable variation. In most cases, however, as comparison will show, there is in the variation an obvious correlation with other variations, notably with that involving the radial canals. But here again the exceptions are sufficiently numerous to warrant the conclusion that there is in this organ itself individual variation, apparently devoid of any adaptive end or relation.

Aside from the facts of meristic nature above noted, there are features of variation which would seem to be of a purely substantive character. For example, in several specimens the manubrium was greatly extended lengthwise, reaching in some cases quite beyond the velum, occasionally as much as one-fourth its total length. While of course this organ is very extensile, yet in many hundreds of specimens examined alive, in many cases while the animal was engaged in engulfing food, I have never seen the manubrium extended beyond the velum.

While no emphasis is placed on this feature of variation, it is yet worthy of note in comparison with such medusae as *Coryne*, *Dipurena*, etc., in which the greatly elongated manubrium feature is rather distinctive.

In Fig. 14 is shown an interesting and anomalous feature which is more or less monstrous, namely, a spike-like growth from one side of the basal portion of the manubrium. While in some respects it might be comparable with an oral tentacle of *Margelis* or *Nemopsis*, still only in a somewhat remote way. It was rather rigid, yet devoid of any chitinous or other rigid support. As will be seen, it has the form of an elongate, attenuate process, about twice the length of the manubrium. It would seem, as suggested above, to be a wholly unique if not anomalous structure, without evident correlation with, or adaptation to, any other organ or function.



FIG. 14. — Semidiagrammatic sketch of manubrium of medusa showing anomalous 'projecting spur, S.

Tentacles.

As compared with *Eucope*, *Obelia*, *Podocoryne*, and many other genera, there seems to be a very different order and relationship among the tentacles of *Gonionemus*. In the smallest specimen measured the diameter was but 2 mm., and the number of tentacles twenty-nine. The largest specimen found measured 19 mm., and the number of tentacles was sixty-eight. That mere size is not, however, determinant of numbers will be seen when it is stated that a specimen measuring 4 mm. had thirty-nine tentacles, while one measuring 6 mm. had but thirty. While, as stated above, the largest measured specimen had sixty-eight tentacles, two others measuring 15 and 16 mm., respectively, had each seventy-two tentacles, and a specimen measuring 14 mm. had seventy-one tentacles. While it should be stated that these observations were made upon specimens preserved in formaldehyde, which may have thereby suffered some shrinkage, still since the preservative was in all cases the same medium and of the same, or very nearly the same, per cent, they would presumably be similarly affected.

However, the matter is not in any wise dependent upon data of this character. Even a glance at Figs. 9-12 will show, though diagrammatically, the relative number and distribution of the tentacles about the margin, while an inspection of the tables will show how very variable is this matter.

Bifurcation of tentacles, tentacular spurs, etc. — In all some fifteen specimens were found having variations involving one or more of the features indicated under this head. As noted by Agassiz and Woodworth ('96) in *Eucope*, the origin of spurs is usually from the base, as is also the doubling of the tentacles, as shown in Pl. IV, Figs. 2, 7, and 10. In several specimens there was an evident bifurcation of the terminal portion, as shown in Pl. IV, Figs. 5, 6, 8, and 9. In the specimens shown in Pl. IV, Fig. 6, this had occurred in close conjunction with the peculiar suctorial bulbs or pads so characteristic of this genus, while in Fig. 8 it is shown as having occurred somewhat proximal to these structures. A single specimen was found having three of these organs on a given tentacle at considerable intervals. In several specimens the tentacular pads or bulbs associated with the bases of the tentacles exhibited peculiar cordate lobings, sometimes on the outer border, more frequently on the inner edge, or from both, as if about to divide, though in no case was division found to be complete in a given bulb. However, this feature will acquire some significance in relation to the double and triple tentacles shown in Fig. 2, where the pads are correspondingly double and triple.

As compared with the specimens of Figs. 7 and 10, however, there will be seen no such correlation,—a fact which would suggest a measure of caution concerning the possible relation of the apparent division of the basal pads and the doubling of tentacles. This caution is further emphasized by the fact that in their origin new tentacles appear wholly apart from these pads, which only after some time are gradually developed on their ventral bases.

I am unable to agree with Agassiz and Woodworth (*op cit.*, p. 139) that these double and triple tentacles are due to coalescence of the bases. Whatever may be the case with *Eucope*,

TABLE I. — TETRAMEROUS SPECIMENS.

RADIAL CANALS.	GONADS.	GASTRIC LOBES.	ORAL LOBES.	TENTACLES.
4	4	4	4	12, 12, 12, 12.
4	4	4	4	12, 12, 15, 16.
4	4	4	4	17, 15, 14, 5.
4	4	4	4	15, 14, 14, 14.
4	4	4	4	12, 11, 13, 13.
4	3	3	3	20, 18, 10, 13.
4	4	4	4	10, 10, 11, 8.
4	4	4	4	16, 15, 17, 17.
4	4	4	4	14, 12, 12, 11.
4	4	4	4	16, 16, 16, 16.
4	4	4	4	14, 14, 14, 14.
4	4	4	4	15, 15, 15, 15.
4	4	4	4	14, 14, 14, 14.
4	4	4	4	12, 12, 12, 8.
4	4	4	4	17, 17, 21, 14.
4	4	4	4	17, 17, 19, 17.
4	4	4	4	16, 16, 16, 16.
4	4	4	4	11, 11, 11, 11.
4	4	4	3	15, 15, 16, 16.
4	4	4	4	12, 11, 12, 10.
4	4	4	4	13, 12, 12, 11.
4	4	4	4	16, 13, 15, 14.
4	4	4	4	15, 15, 15, 15.
4	3	4	4	13, 13, 10, 9.
4	4	4	4	13, 11, 12, 12.
4	4	4	4	11, 11, 11, 11.
4	4	4	4	12, 10, 11, 11.
4	4	4	4	13, 14, 14, 13.
4	4	4	4	17, 17, 17, 15.
4	4	4	4	14, 14, 14, 14.
4	4	4	4	15, 16, 14, 15.
4	4	4	4	16, 15, 16, 14.
4	3	3	3	12, 11, 8, 4.
4	4	3	3	11, 12, 11, 11.
4	4	4	4	12, 12, 13, 13.
4	4	5	5	21, 13, 13, 14.
4	4	4	4	11, 14, 11, 7.
4	4	4	4	14, 14, 13, 17.
4	3	4	4	14, 15, 14, 9.

TABLE II.

RADIAL CANALS.	GONADS.	GASTRIC LOBES.	ORAL LOBES.	TENTACLES.
3	3	3	3	14, 20, 22.
3	3	3	3	14, 19, 19.
3	3	3	3	20, 21, 21.
3	3	3	3	16, 20, 22.
3	3	3	3	14, 18, 24.
3	3	3	3	15, 15, 17.
3	3	2	2	13, 17, 24.
3	3	3	3	12, 15, 18.
3	3	3	3	13, 15, 18.
3	3	3	3	12, 12, 21.
3	3	4	4	13, 14, 18.
3	3	3	3	15, 15, 16.
3	3	3	5	15, 17, 23.
*4	4	4	4	9, 13, 16.
*4	4	4	4	15, 15, 17.
5	5	4	4	11, 12, 14, 17, 18.
5	4	4	4	8, 12, 14, 15, 16.
5	5	5	5	7, 8, 9, 10, 11.
5	5	4	4	11, 9, 9, 11, 8.
5	5	3	3	4, 9, 11, 9, 15.
5	5	4	4	12, 10, 7, 8, 13.
5	5	5	5	12, 11, 11, 7, 11.
5	5	4	4	12, 10, 10, 11, 11.
5	5	4	4	11, 7, 9, 9, 11.
5	5	5	5	8, 9, 7, 9, 8.
5	5	4	4	6, 10, 14, 12, 10.
5	5	5	5	11, 13, 9, 10, 11.
5	5	5	5	8, 9, 15, 17, 16.
5	5	4	4	11, 10, 13, 13, 13.
6	6	5	5	6, 5, 6, 6, 6, 6.
6	6	4	4	7, 9, 8, 12, 18, 9.
6	6	6	5	7, 9, 8, 6, 4, 8.
6	6	6	5	7, 11, 7, 8, 9, 5.
6	6	5	5	11, 4, 9, 7, 7, 12.

* While in these specimens four canals were present, two in each were so closely approximated as to divide the bell into trimerous sectors.

with *Gonionemus* all the facts would seem to point to their independent origin, in many cases the tentacles being of conspicuously different sizes, and in others exhibiting all phases of intermediate conditions between the simple bifurcation of the terminal portion through the budding of a smaller tentacle from the side of the larger, on to the symmetrical double tentacles as shown in Figs. 3, 4, and 10.

In only one case was a trifid tentacle found. This is shown in Pl. IV, Fig. 9. However, while a trifid structure there seems to be a degeneration of the median lobe, which was in all probability the terminal portion of what was originally a normal tentacle, from which later were budded the two lateral shoots, each in turn becoming more prominent than the median tip and developing in the appropriate places the characteristic suctorial pads. The degenerating middle tip would very naturally suggest the probability that an injury might have been the predisposing cause of the secondary tips; on the other hand, it must not be overlooked that in each of the other specimens with double tips no such cause seems at all evident. I am inclined to consider the cases as simply the expression of the intrinsic capabilities of variation, more or less evident in the several classes of organs already considered.

As yet no reference has been made to the order in which second, third, and subsequent series of tentacles arise. *Gonionemus*, unlike *Eucope*, seems to have no such association of tentacle and sensory bulb as serves to locate in part the primary set of tentacles and the order of appearance of the subsequent series. While usually there is a single primary tentacle at the terminus of each radial canal, this is not invariably the case. An inspection of Figs. 9-12 will show that there may be a very wide range of variation in this respect. The following data, taken at random from many observations, will further illustrate the same general fact.

the bell the adequate marginal space becomes available. And the fact that this seems so variable would appear to warrant the inference that growth occurs at irregular intervals and areas over the body of the medusa. Might not this fact throw some light upon the marked unsymmetry of such forms as those shown in Figs. 10 and 11? This suggestion would seem to find some further support in the fact that very young specimens appear to be more constant in their symmetry than those more mature.

Otocysts.

In formalin specimens there is a degree of opacity induced, especially about the marginal area of the bell, which often renders difficult any satisfactory examination of these sensory bodies. Hence only a limited number of critical determinations on this point were made, but these were sufficient to show a degree of variation both in their number and arrangement quite as marked as in that of other organs.

Normally they should occur in somewhat symmetrical order between the bases of the tentacles. This, however, is rarely the case. There seems about the same variation in their occurrence and relations as in the case of the tentacles, though I was not able to discover that the latter had any determining influence upon them. In only a few cases have I been able to demonstrate the presence of more than a single otolith in a given cyst, and in no case more than two. On this point, however, the opacity above referred to, and the abundant pigment about the bases of the tentacular bulbs were material obstructions to such determinations, and suggest tentative conclusions. In matter of shape and size these organs present likewise considerable variation. Pl. IV, Fig. 1, presents the average aspect of form at *a*, while at *b* are shown forms not unusual but variant.

Summary and Review.

1. Variation among Hydromedusae is of wider extent than had been supposed.
2. Variation is much greater in some genera than in others.

3. Variation among Hydromedusae appears to be much less symmetrical and less definitely correlated than among Scyphomedusae.

4. Many phases of variation seem to be wholly devoid of any adaptive features or tendencies.

5. The ratio of variation is higher among the tentacles than among other organs, and in many species higher than in all other organs combined, — a feature which is perhaps the most conspicuous case of adaptation apparent in the entire series.

Among the earliest references to variation in Coelenterates is that of Ehrenberg ('37) relative to variation in Aurelia. Later, Romanes ('74-'76) took up the subject with much more detail, giving an extended account of the nature and extent of variation, particularly in Aurelia, in which he figures and describes many "monstrous forms of medusae" and points out interesting correlations of radial canals, gonads, tentacles, etc.

Within recent times these observations have been much extended, notably by Brown ('94), who distinguished more than two per cent.

Sorby ('94), Herdman ('94), and Unthank ('94) have each recorded many interesting facts of variation in this medusa.

In 1895 Brown still further extended his observations upon Aurelia, and in connection therewith undertook a comparison of a large number of the Ephyrae. He was able to distinguish no less than 22.6 per cent of numerical variation in tentaculocysts, a ratio very close to that earlier determined for adult Aurelia. The observations seemed to show upon the whole a tendency toward an increase in meristic characters.

Ballowitz ('98) records extended observations upon Aurelia, specially with reference to the gonads. While in general there was more or less correlation in the numerical variation of these organs with the actinal lobes, it was apparently less constant than had been claimed by earlier observers. The highest number noted was seven, while three was the minimal number. One specimen in particular, which he names Ephyra abnormitat, seems to be an unusual monstrosity, having a very large balloon-shaped body with a correspondingly large manubrium.

He explains it as probably due to an enormous expansion of the top of the Ephyra, thus forming the balloon-like body.

It may not be amiss in this connection to record observations of a similar character as to numerical variations upon the Aurelias of Woods Holl which quite confirm those cited.

Observations upon the Hydromedusae seem to have been heretofore quite limited. Those of Forbes ('48), Agassiz ('49), Hincks ('68), Romanes ('74-'76), Agassiz and Woodworth ('96), include all the more important observations which have come to my knowledge. The latter would seem to be about the only series made upon a large number of specimens with the purpose of ascertaining the extent and character of variation in a single genus.

SYRACUSE UNIVERSITY, September 1, 1900.

REFERENCES.

- AGASSIZ, A., and WOODWORTH. *Bull. Mus. Comp. Zool.* Vol. xxx. No. 2. 1896.
- BALLOWITZ. Variation in Aurelia. *Arch. f. Entwickl. d. Organismen.* 1898.
- BATESON, W. Materials for Study of Variation. p. 424 *et seq.* 1894.
- BROWN, E. T. Variation of Tentaculocysts of Aurelia aurita. *Quar. Journ. Micr. Sci.* Vol. xxxviii.
- BROWN, E. T. Variation of Haliclystus octoradiatus. *Quar. Journ. Micr. Sci.* Vol. xxxviii.
- BROWN, E. T. Variation of Aurelia. *Nature.* Vol. l. 1894.
- FORBES, EDW. British Naked Eye Medusae. London, 1848.
- HARGITT, C. W. Nat. Hist. of Pennaria. *Amer. Nat.* May, 1900.
- HERDMAN, W. H. Pentamerous Aurelia. *Nature.* Vol. l. 1894.
- HINCKS, T. Clavatella. *Hydroid Zoöphytes.* p. 71. 1868.
- HORNELL, J. Abnormalities in Haliclystus octoradiatus. *Nat. Sci.* 1893.
- HORNELL, J. Lucernaria as Degenerate Scyphomedusae. *Nat. Sci.* 1893.
- MURBACH, L. *Journ. of Morph.* 1895.
- ROMANES, J. G. Varieties and Monstrous Forms of Medusae. *Journ. Linn. Soc.* 1874-76.
- SMALLWOOD, M. Morphology of Pennaria. *Amer. Nat.* 1899.
- SORBY, H. C. Symmetry of Aurelia aurita. *Nature.* Vol. l. 1894.
- UNTHANK, H. W. Pentamerous Aurelia. *Nature.* Vol. l. 1894.

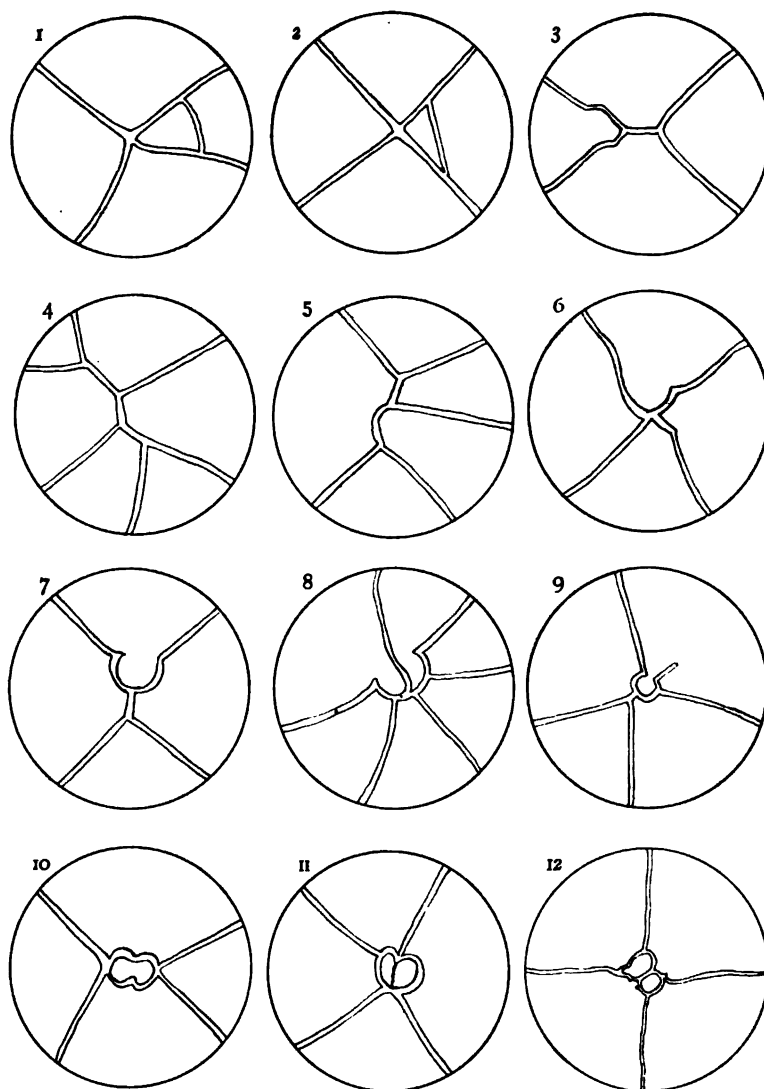


PLATE I. — Diagrammatic figures, illustrating variation in form, number, and arrangement of the radial canals.

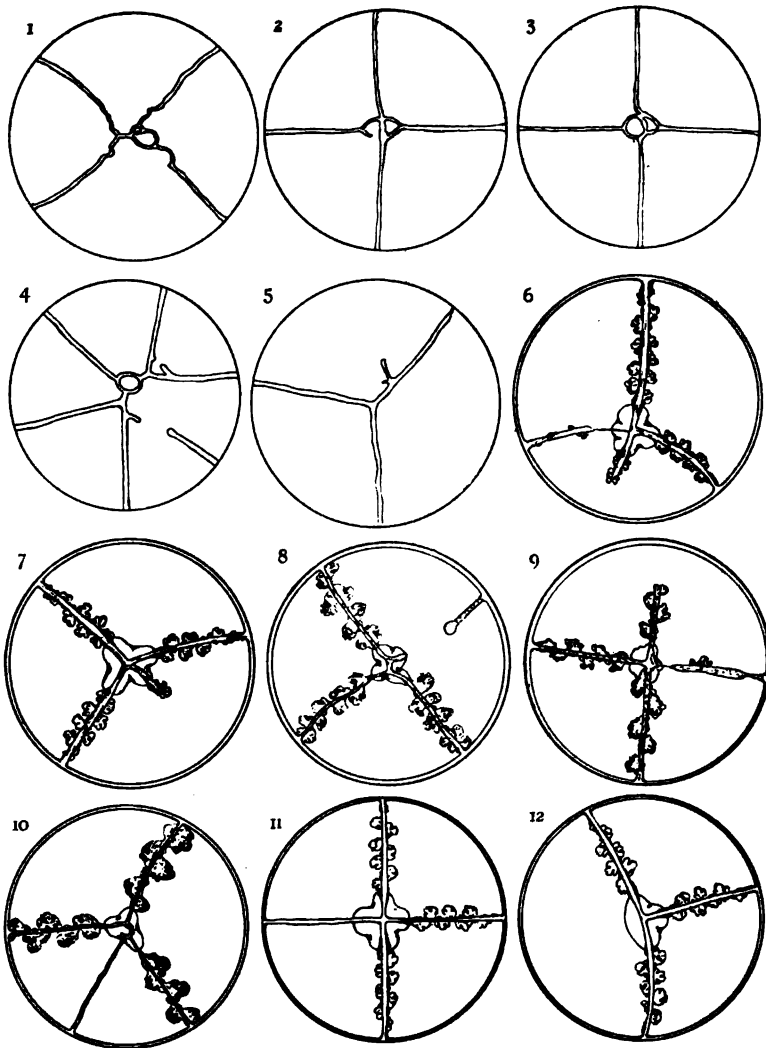


PLATE II. — Showing various phases of atrophy, spur-like branches, etc. of radial canals.

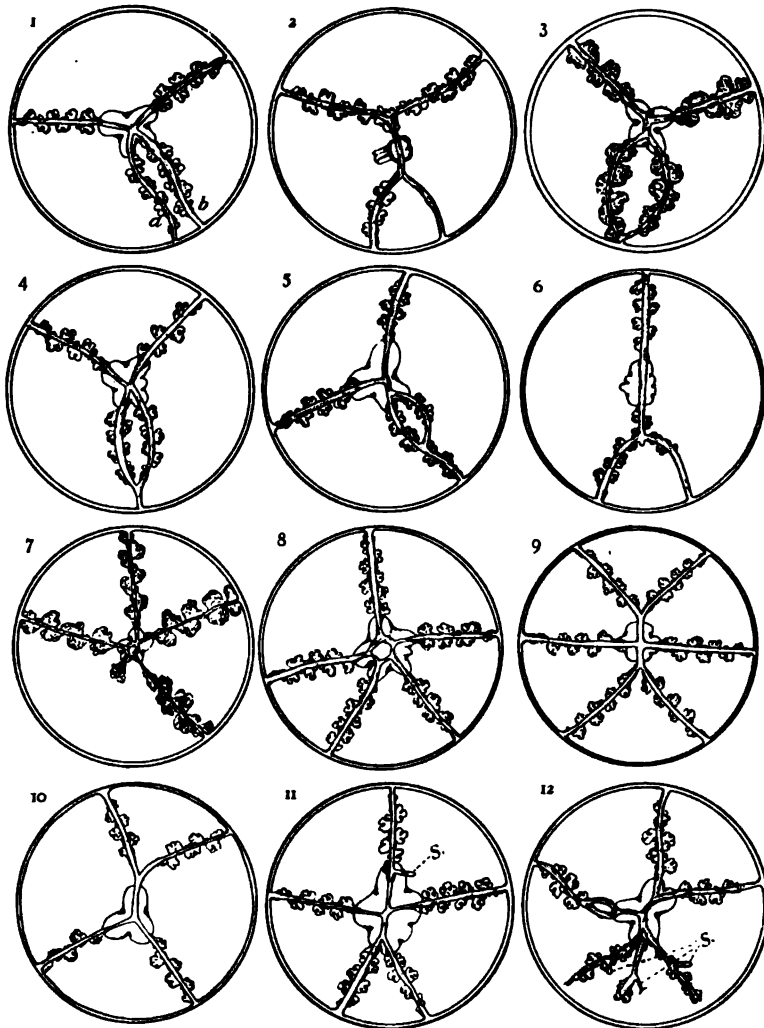


PLATE III. — Figs. 1-6 show varying phases in the evolution of trimerism. Figs. 11 and 12 show at *S* the development of spurs.

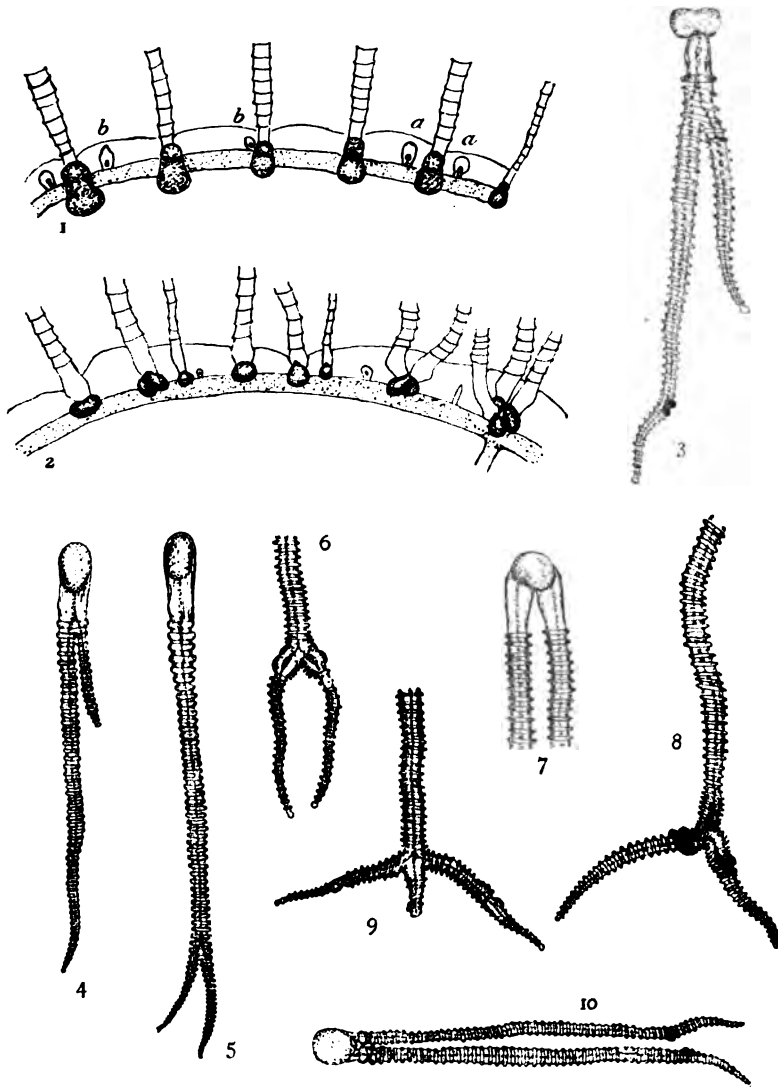


PLATE IV. — Fig. 1, *a, a*, normal; *b, b*, variable forms of otocysts. Fig. 2 showing variation in tentacles. Figs. 3-10 show various phases in forking or budding of tentacles.

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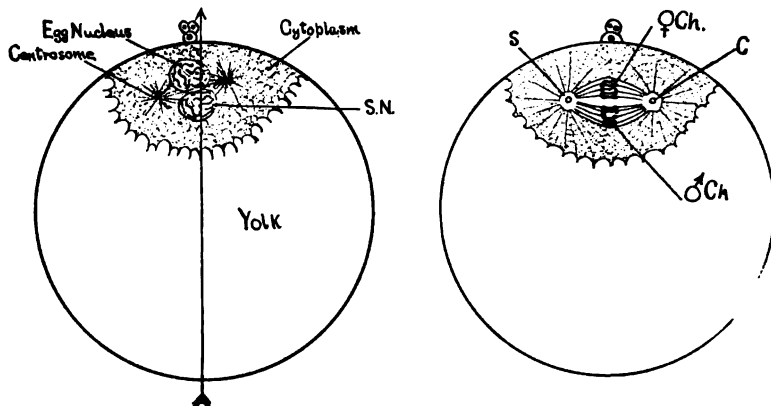
[From the Zoölogical Laboratory of the University of Pennsylvania.]

THE INDIVIDUALITY OF THE GERM NUCLEI DURING THE CLEAVAGE OF THE EGG OF CREPIDULA.

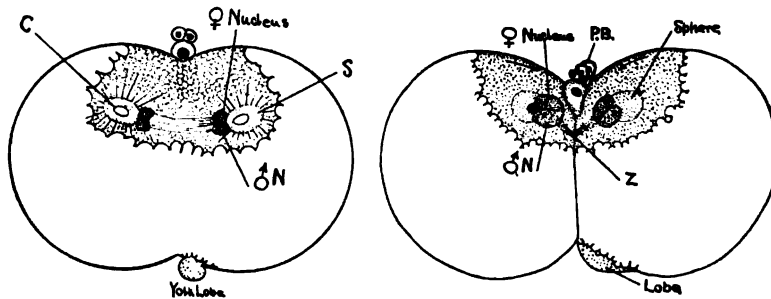
EDWIN G. CONKLIN.

HAECKER ('92) and Rückert ('95) have made known the interesting fact that the germ nuclei of *Cyclops* do not fuse but preserve their individuality throughout a considerable portion of the cleavage of the egg. Herla ('93) and Zoja ('95) have shown that the paternal and maternal chromosomes of *Ascaris* remain distinct at least as far as the 12-cell stage. These observations are of the greatest significance and, so far as they go, establish Boveri's hypothesis ('91), "that in all cells derived in the regular course of division from the fertilized egg, one-half of the chromosomes are of strictly paternal origin, the other half of maternal." So far as I am aware, similar observations have not hitherto been made in the case of other animals.¹

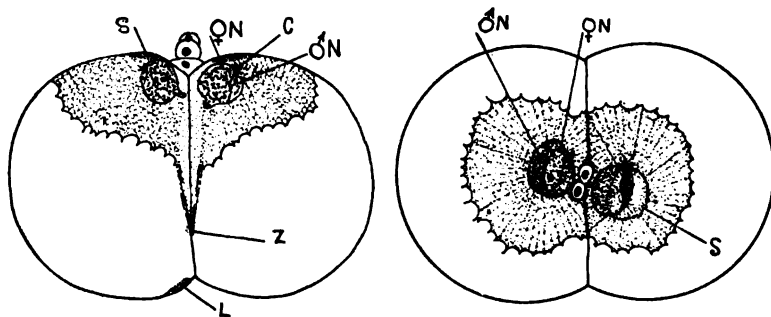
¹ Rückert calls attention to the fact that partially cleft nuclei are found in the figures of various authors, particularly those of Fol ('79) on *Toxopneustes*, of Bellonci ('84), and of Kölliker ('89) on *Siredon*. Of course no one of these observers has interpreted these figures as showing the independence of the germ nuclei, and some of the figures referred to by Rückert probably do not show this phenomenon. For example, only one of Fol's figures (Pl. VII, Fig. 7) shows a dual nucleus, while the figure in Kölliker's text-book (Fig. 36) is probably a case of the indentation of the nuclear membrane opposite the centrosomes in the early prophase, a thing which frequently happens. Bellonci's Figs. 1 and 20 show an indentation on one side of the nucleus which may correspond to a division between the germ halves, though this must be regarded as more or less doubtful.



FIGS. 1 and 2. — Prophase and Metaphase of First Cleavage. S.N. = Sperm Nucleus. Ch. = Chromosomes. S. = Sphere. C. = Centrosome.

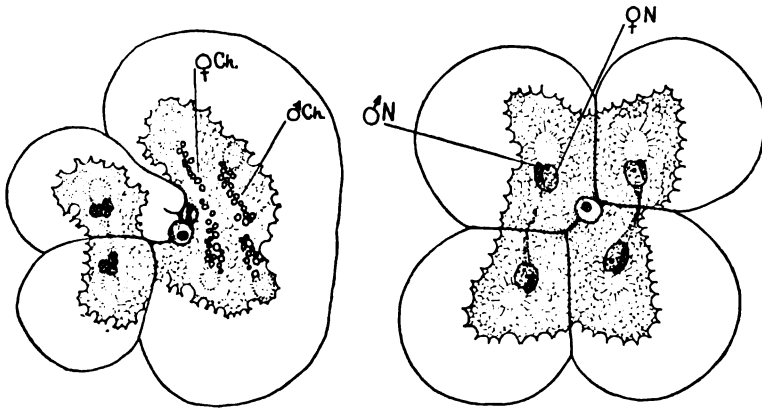


FIGS. 3 and 4. — Anaphase and Telophase of First Cleavage, showing dual nuclei, centrosomes (C) and spheres (S), "Zwischenkörper" (Z), bending of spindle axis, and progressive absorption of yolk lobe.



FIGS. 5 and 6. — Prophase of Second Cleavage, showing dual nuclei with central spindle lying in groove between the halves; Fig. 5 viewed from one side, Fig. 6 from animal pole. The spheres (S) lie over the nuclei and immediately under the cell wall; the spindle axis is bent on itself, and the "Zwischenkörper" (Z) is carried nearly to the vegetal pole; the nuclei shows processes projecting toward the "Zwischenkörper," and the yolk lobe (L) is almost completely absorbed.

In *Crepidula plana* I have observed a separateness of the germ nuclei in certain stages of the nuclear cycle which is of such a character that it may lead to the discovery of similar phenomena in other animals. This separateness is most easily



FIGS. 7 and 8. — Anaphase and Telophase of Second Cleavage. Fig. 7, an abnormal egg in which the left half has divided normally and the chromosomal vesicles of the daughter-nuclei are fusing; in the right half the division figure is double with four spheres and two groups of chromosomal vesicles which have not fused; there are thirty chromosomal vesicles in each group. Fig. 8, a normal egg showing the egg and sperm constituents in each of the daughter-nuclei.

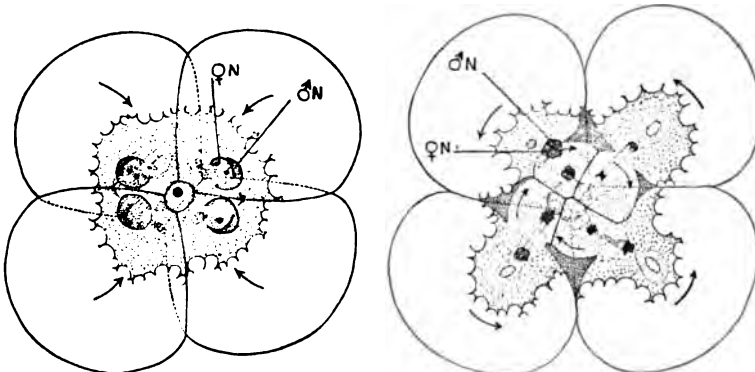


FIG. 9. — Telophase of Second Cleavage; centrosomes, spheres, and nuclei rotating in direction of arrows.

FIG. 10. — Anaphase of Third Cleavage. Egg and sperm constituents of nuclei indicated.

observed in the telophase of each division, though in some cleavage cells it may be seen in the prophase also, or even throughout the resting period. At the time when the daughter-nuclei are being formed the chromosomal vesicles fuse into two

groups which are closely pressed together but are still separated by a partition wall (Figs. 4, 8, *et seq.*), as Rückert has shown to be the case in Cyclops. Gradually this partition wall disappears, being preserved longest on that side of the nucleus nearest the centrosome (Fig. 5). Here a groove is formed on one side of the nucleus which marks the line of contact between the two halves. In some cleavage cells this groove is visible throughout most of the resting period (Figs. 4, 9); in others it disappears during the greater part of the resting period, though it may reappear in the following prophase (Figs. 5, 6); in all cases, however, the partition wall and groove reappear in the next succeeding telophase, when it is formed again in the manner described above. I have observed the double character of the nucleus in the telophase of every cleavage up to the 29-cell stage (Figs. 1-16), and in several of the later cleavages up to the 60-cell stage, and I have no doubt that it is found in all the later cleavages, though it becomes more difficult to see as the nuclei grow smaller. While the halves of these double nuclei occupy similar positions relative to each other at corresponding stages in any cell generation, they occupy different positions at different stages and in different generations; consequently the position of the groove or partition wall which separates the halves of the double nuclei can be satisfactorily studied only in preparations of entire eggs, which may be observed from all sides. All the figures which illustrate this paper are therefore of entire eggs, though many isolated cases of double nuclei have been observed and studied in actual sections.

On each side of the partition wall which divides these double nuclei there is usually a single small nucleolus; these two nucleoli persist long after the disappearance of the partition and frequently throughout the whole of the resting period. In most if not all of the early cleavages there are two, and only two, nucleoli present in the telophase (Figs. 3, 4); but if this is succeeded by a very long resting period the number may increase to more than two, or all may fuse into a single enormously large one.

It still remains to show that these double nuclei really

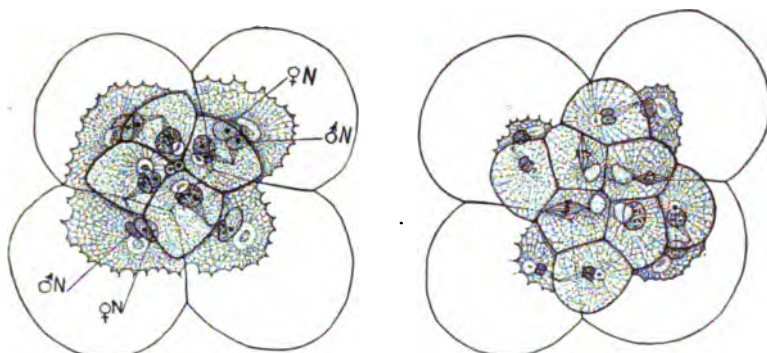


FIG. 11. — Telophase of Third Cleavage. Egg and sperm constituents of nuclei indicated; also bending of spindle axis and rotation of centrosomes and nuclei.

FIG. 12. — Telophase of Fourth Cleavage and Prophase of division of First Quartette cells. The nuclei in the telophase are dual, though from this stage on the egg and sperm constituents cannot be identified with certainty.

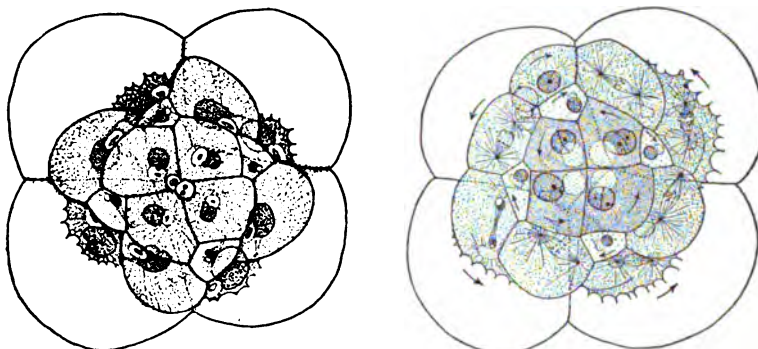


FIG. 13. — Telophase of Division of First Quartette. Dual nuclei in each daughter-cell.

FIG. 14. — Subdivision of Second Quartette and formation of Third. Dual character of nuclei shown in apical cells.

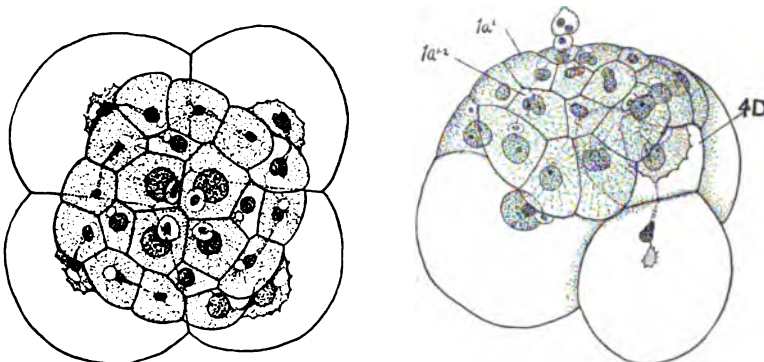


FIG. 15. — Telophase of Division of Second Quartette and of formation of Third; dual nuclei shown in almost all of these cells.

FIG. 16. — Telophase of formation of the Mesentoblast Cell (4D) and of the second division of the First Quartette (1a¹, 1a¹·², etc.); dual nuclei shown in both cases.

represent the egg and sperm nuclei which have not yet lost their individuality. This cannot be demonstrated in *Crepidula*, for the reason that this double character is not apparent at every stage in the nuclear cycle, but it is extremely probable, as the following observations will show :

1. In the first cleavage the germ nuclei do not fuse but remain distinct throughout the prophase, and even in the metaphase they are represented by separate groups of chromosomes (Figs. 1, 2) ; in the early anaphase these groups of chromosomes can no longer be distinguished, though I think they must still remain separate, for the nuclei are clearly double in the immediately following late anaphase and telophase (Figs. 3, 4). The position of the partition wall in these double nuclei corresponds to the plane of contact between the germ nuclei; the egg nucleus always lies more or less above the sperm nucleus, and in the telophase of the first cleavage one-half of each double nucleus overlaps the other half to a greater or less extent (Figs. 1-4). It is probable that the upper half represents the egg nucleus, and the lower half the sperm nucleus, and in all the later cleavages it is probable that the half of the nucleus which lies nearest the animal pole is from the egg, and the other half from the sperm.

2. The groove which is found on one side of the nucleus in the telophase of the first cleavage (Fig. 4) persists well into the resting stage, and a corresponding groove is found in the same position in the prophase of the second cleavage (Figs. 5, 6). The central spindle for the second cleavage lies in this groove (Fig. 6), and thus the amphiaster actually lies in the only plane in which it would be possible to halve the two parts of the double nuclei. This very fact shows that each half of a double nucleus is represented in the daughter-nuclei, and it strongly suggests that the two parts of the daughter-nuclei are derived directly from the corresponding parts of the mother-nucleus (*cf.* Figs. 6, 8). The fact that the central spindle lies in the groove separating the halves of the nucleus has been observed in the first, second, third, and fourth cleavages, and is undoubtedly a general phenomenon. There is no reasonable ground for doubting that the two parts of every double nucleus are

derived from corresponding parts of a mother-nucleus, and so on back to the egg and sperm nuclei in the first cleavage.

Since the descendants of the germ nuclei are halved at every division, it follows that successive divisions of the double nuclei cannot be at right angles to one another, since this would lead to an unequal division of the halves, or even to a division along the plane of contact between the halves. Such an unequal division might be prevented in cleavages which successively alternate in direction by the rotation of the nucleus during the resting period, or by the rotation of the spindle in the early stages of mitosis.¹ As a matter of fact both of these methods occur in *Crepidula*. The nucleus usually rotates during the rest through 90° , so that although successive nuclear spindles are at right angles to one another the axis of every spindle lies in the same nuclear axis (*cf.* Figs. 3, 4, 8, 9); but in some cases the nuclear spindle does not lie in its definitive position when first formed but undergoes extensive rotation after its formation. While it is not susceptible of absolute proof, since the partition wall is absent during the later stages of the rest, it is highly probable that the plane in which all nuclear spindles lie is the plane of contact between the two halves of every nucleus.

3. In certain abnormal cleavages the double nuclei are really two entirely separate nuclei lying side by side within a single cell. Such binucleated cells may occasionally be found with the nuclei in the height of the rest, though they are more usual in the telophase or early resting period. There is usually but a single sphere and centrosome in such cells, though in one case of pathological mitosis which I have seen (Fig. 7) there are two mitotic figures side by side; the chromosomes which have reached the stage of the chromosomal vesicles have not aggregated at the poles of these spindles, but are scattered along their whole length. There are thirty of these

¹ Rückert finds that the nuclei rotate in *Cyclops* even after the spheres have reached their definitive positions at the poles of the spindle; I have never observed in *Crepidula* a rotation of the nuclei, independent of the spindles, at so late a stage in the cell cycle.

chromosomes in each spindle, the same number that is found in each germ nucleus.

4. In each of the germ nuclei, before they come into contact, there is a single nucleolus; these nucleoli disappear in the prophase of the first cleavage, but in the succeeding telophase a single nucleolus generally appears in each half of each daughter-nucleus. The same is true of the succeeding cleavages, so that each nucleus throughout the cleavage usually has two nucleoli in the telophase or early resting stage, though the number may vary in the later resting period, as pointed out above. The fact that there is a single nucleolus in each germ nucleus, and that there is usually a single nucleolus in each half of the double nuclei of the cleavage, may possibly indicate that these halves are each derived from one of the germ nuclei. Since the nucleoli as such do not persist throughout the mitosis, may it not be possible that there is some achromatic structure in connection with them which does persist and form the basis for the new nucleoli which appear in the daughter-nuclei?

These facts make it very probable that the germ nuclei of *Crepidula* preserve their individuality throughout the cleavage, though their separateness may be apparent only or chiefly in a single stage of the nuclear cycle, *viz.*, the telophase. Further, it is possible, even in an advanced stage of the cleavage, to determine with considerable probability which part of a double nucleus is derived from the egg and which from the sperm, the egg half always lying nearer the animal pole than the sperm half. Finally the initial position of the mitotic spindle seems to be determined by the relative positions of the halves of the double nuclei, since the spindles when they first appear lie in the plane of contact between the two halves; the final position of the spindle and the direction of division are determined by the movements of the cytoplasm.

REFERENCES.

- '84 BELLONCI, G. Interno alla cariocinesi nella segmentazione dell' ovo de Axolotl. *Atti della Accad. dei Lincei*, Memoire xix.
- '79 FOL, H. Recherches sur la fécondation et le commencement de l'henogenie. Geneve.
- '92 HÄCKER, V. Die Eibildung bei Cyclops und Canthocamotus. *Zool. Jahrb.* Bd. v.
- '93 HERLA, V. Étude des variations de la mitose chez l'ascaride megalocephale. *Arch. Biol.* Vol. xiii.
- '89 KÖLLIKER, A. Handbuch der Gewebelehre. 6 Aufl. Leipzig.
- '95 RÜCKERT, J. Ueber das Selbständigbleiben der väterlichen und mütterlichen Kernsubstanz während der ersten Entwicklung des befruchteten Cyclops-Eies. *Arch. f. mikr. Anat.* Bd. xlv.
- '95 ZOJA, R. Sulla indipendenza della cromatina paterna e materna nel nucleo delle cellule embryonale. *Anat. Anz.* Vol. xi.

THE EARLY DEVELOPMENT OF THE HYPOPHYSIS IN CHELONIA.

CHARLES ZELENY.

THE following observations on the early development of the hypophysis in Chelonia are offered at this time because they throw positive light upon the derivation of the organ concerning which we at present have several conflicting views. The paper is based upon sections of *Aspidonectes spinifer* Ag., *Chelydra serpentina* L., and *Chrysemys marginata* Ag., which show that in these forms the hypophysis is undoubtedly of epiblastic origin. Incidentally some points regarding the relation of the preoral gut to the notochord and the head cavities will be noted, but this subject will receive fuller treatment in a subsequent paper. For the sake of convenience the subject matter will be considered under the following heads :

1. A general outline of the literature dealing with the early development of the hypophysis among vertebrates.
2. Material and methods of preparation.
3. Description of stages.
4. Summary and conclusion.

Literature.

It would be out of place in a paper such as the present to give any detailed account of the views which have been held regarding the subject under discussion. A bare mention of a few of the upholders of each of the principal views, giving the group upon which work was done, must suffice.

1. Those who have claimed a hypoblastic origin for the hypophysis are Luschka ('69), Mammalia ; W. Müller ('71), Vertebrata in general ; Dohrn ('82), Teleostii, later extended to Vertebrata in general ; Hoffmann ('88), Lacerta ; Prather ('99), Amia.

2. Those who have claimed an epiblastic origin for the hypophysis are Mihalkovics ('77), Aves and Mammalia ; Balfour ('78), Elasmobranchii ; Orr ('87), Lacerta ; Lundsberg ('94), Salmonidae ; Dean ('96), Amia ; Haller ('96), Vertebrata in general ; Hoffmann ('96), Elasmobranchii ; Melchers ('99), Lacertilia. Of these Orr ('87), although he describes the hypophysis as of epiblastic origin and so figures it in his sections, nevertheless considers it probable that hypoblast cells may take some part in its development.

3. Those who have claimed that the hypophysis is partly of epiblastic and partly of hypoblastic origin are Kupffer ('93), Acipenser and Ammocoetes ; Valenti ('95), Amphibia (Bufo) ; Nussbaum ('96), Mammalia ; and considered probable by Orr ('87), Lacerta.

It is of special interest to note that even this partial list gives each of the three views a large number of the groups of vertebrates upon which to base its general character.

Material and Methods.

The material upon which the following observations are based was obtained in Minnesota during the summers of 1898 and 1899. The embryos of *Aspidonectes* are from Grey Cloud Island in the Mississippi River below St. Paul, and those of *Chrysemys* and *Chelydra* are from the neighborhood of Hutchinson. The fixing fluid used was Gilson's mercuronitric mixture. The embryos were stained *in toto* in haemacalcium or in paracarmine. The series of sagittal sections were found to be the most helpful in determining the cell-layer from which the hypophysis is derived, and the following descriptions are taken entirely from such sections.

Description of Stages.

The following stages will be figured and described :

Stage A. The hypophysial evagination has not yet appeared.

Stage B. The hypophysial evagination is very evident and the pharyngeal membrane has not yet been broken.

Stage C. Slightly older than Stage B. The pharyngeal membrane has been broken.

Stage D. The downward growth and enlargement of the fore-brain have pushed the hypophysis back from its primary relation to the broken ends of the pharyngeal membrane.

Stage E. The hypophysis has been shifted backward so as to assume a position relatively far back in the pharyngeal cavity.

Stage F. The hypophysis has become differentiated into a terminal, broad, sac-like part and a narrower connecting stalk.

Stage A.

In an embryo with five or six mesoblastic somites and in which the medullary folds have not yet united above to enclose a medullary canal the hypophysial evagination has not begun to form. A median sagittal section of such an embryo, however, shows the relation of the parts surrounding the point at which the hypophysis will appear at a later stage. Fig. 1, Pl. I, represents a diagrammatic median sagittal section of an embryo of *Chrysemys marginata* 2.5 mm. in length and in which there are five distinct mesoblastic somites with indistinct traces of a sixth. The figure is a combination of the six sections nearest to the median line. Although the medullary groove is still open above, the medullary folds in the head region have grown to a considerable height, as represented by the dotted line *D*. At the same time the whole anterior region has been folded and bent downward. The fold of the blastoderm which comes up over the head as a result is the proamnion (*Pa.*), and consists of epiblast and hypoblast. At the bottom of the head fold the hypoblast has traveled back much farther than the epiblast, leaving a space in which the cardiac mesoblast (*Cm.*) develops. As we trace this hypoblast (*End.*₁) forward along the floor of the fore-gut (*F.G.*) we find that it is bent ventrally so as to come into contact with the epiblast. This point of contact, represented by the double-headed arrow (↑), is the region at which the mouth will be formed. In front of this we may recognize a short, wide, preoral gut (*Pr.G.*). This is a

true preoral gut and not an apparent one caused by the downward bending of the head region; for we must consider the sharp angle in the hypoblast at *Pr.G.* in the figure, and not the part back of it where the mouth will appear, to have been the original extreme anterior end. Thus we may consider all the hypoblast below and posterior to this angle to belong to the ventral wall of the alimentary canal, and all that above and posterior to belong to the dorsal wall. Following the dorsal wall we find that it immediately divides into two parts, one of which is the notochord (*Nc.*) and the other the dorsal wall of the gut (*End.*₂). For this reason the hypoblast forming the ventral wall, of the alimentary canal, and continued out at the ends of the embryo into the flat outlying blastodermic region, may be called the "primary hypoblast," and the hypoblast of the dorsal wall the "secondary hypoblast." Rex ('97), in his work on the duck, and Davidoff ('99), in the embryo of *Platydictylus*, have found similar relations of the notochord to the hypoblast. The terms "primary hypoblast" and "secondary hypoblast," which are used above, are taken from the paper of the latter author. It is important to note in this connection that my sections show a distinct line of demarcation between the epiblast and the hypoblast in this region, so that the mass of cells surrounding the preoral gut is distinctly hypoblastic and not a mass of undifferentiated cells. The arrow with a feathered shaft () shown in Fig. 1 in the epiblast directly under this point marks the position and direction of the future hypophysial pocket. The very plain line of division between the epiblast and hypoblast excludes the possibility that any of the hypoblast cells may take part in this ingrowth of the epiblast.

Stage B.

In an embryo 5.2 mm. in length and with twenty-one mesoblastic somites, such as is shown in median sagittal section in Pl. I, Fig. 2, and Pl. II, Fig. 3, the brain has developed with great rapidity. The cephalic flexure having proceeded at the same time, the region around the preoral gut is greatly compressed. The cavity of the preoral gut itself has become very

small and its dorsal wall is doubled back on itself. The notochord also at its anterior end near the point where it joins the hypoblast has become very much twisted and curved, giving it a knotted appearance in sagittal section. However, the parts are easily recognized as having the same relation as those in Stage A (Fig. 1). In Stage B (Fig. 2), as before, the dorsal wall of the fore-gut is the "secondary hypoblast" and the ventral wall the "primary hypoblast," the division line between the two being the point where the notochord joins the hypoblast. The pharyngeal membrane is still intact but breaks up very soon after this stage. Directly in front of the place where the mouth opening will appear the epiblast bends sharply back on itself to follow the brain, forming a pocket in which the cells are taller than in the neighboring parts of the epiblast. This is the beginning of the outpocketing which will eventually form the hypophysis. The evaginating layer of cells is clearly distinct from the hypoblast cells of the preoral gut, as is well shown in Pl. II, Fig. 3, which gives an enlarged view of the hypophysial region of Fig. 2. Starting from the condition of the stomodeal epiblast in the earlier stage as shown in Fig. 1, we see that an epiblastic groove was originally formed at the point marked by the feathered arrow (↗) just in front of the future mouth by the forward and downward bending of the fore-brain. It is at the bottom and in the middle of this groove that the thickening and outpocketing of the cells start, and later form the epiblastic pouch which becomes the oral portion of the hypophysis.

Stage C.

In an embryo but slightly older than the one last described the pharyngeal membrane has already been broken. Fig. 4, Pl. III, represents a diagram of a median sagittal section of such an embryo. On account of a lateral bend in the neck region it was not possible to obtain a section which would show the connection of the notochord and the preoral gut at the same time with the mouth opening and the hypophysial evagination. The canal (*H.C.*) between the two premandibular

head cavities is, however, shown, and the small mass of cells connected with it and directed toward the hypoblast of the fore-gut is the strand which connects the head cavities with the preoral gut. The preoral gut itself is not shown in this figure, the cavity (*F.G.*) being a part of the fore-gut which has assumed a position anterior to the mouth because of the bending of the alimentary canal, which takes place at the same time with the cephalic flexure. Except for the break in the pharyngeal membrane the relation of the hypophysis to the epiblast is the same as in Stage B. The hypophysial outpocketing is here, as before, on the epiblastic side of the mouth opening and is undoubtedly made up entirely of epiblast cells.

Stage D.

In Fig. 5, Pl. III, we have a median sagittal section of a somewhat later stage. The points *p* and *p'* show the position of the ends of the broken pharyngeal membrane, and the dotted line (*P.M.*) between them represents the former position of the now ruptured membrane. The hypophysial pouch (*Hyp.*) is shown very distinctly on the epiblastic side of the membrane. It has, however, been pushed back from its original position with relation to the point *p'* by the rapid growth of the fore-brain. The anterior end of the notochord (*N.*) is still more curved and wrinkled than in the last stage, but it retains its connection with the anterior wall of the preoral gut (*P.G.*) by means of a string of cells. In this string of cells we see the section of the canal (*H.C.*) which connects the two anterior or pre-mandibular head cavities. The hypophysial evagination from the very beginning is in close contact with the wall of the infundibulum, but the two layers of cells always remain clearly distinct. The epiblast cells of the hypophysis also remain clearly distinct from the hypoblast cells of the preoral gut.

Stage E.

Fig. 6, Pl. IV, is a diagram of a section of *Chelydra serpentina*. It shows a continuation of the same process of enlargement of the fore-brain and the consequent pushing

back of the hypophysial evagination, so that the latter finally appears to lie far back in the pharyngeal cavity. However, here, as before, the dotted line *pp'* shows the original position of the now broken membrane. At this stage the notochord has severed its connection with the fore-gut, but it is still joined to the mass of cells which connects the two head cavities. These cells still surround a canal, so that there is a passageway from the premandibular cavity on one side of the head to the corresponding cavity on the other side.

Stage F.

Fig. 7, Pl. IV, represents a median sagittal section of *Aspidonectes spinifer*, and Figs. 8 and 9, Pl. V, represent sections of the same series, respectively, three and six sections to the side of the median line. In these it is seen that there is no evidence of the canal which connected the two head cavities, and the notochord shows no sign of the former union with the hypoblast. The hypophysis has begun to constrict in the basal region and to enlarge in the terminal region so as to show a division into a narrower basal stalk and a wider terminal sac-like portion.

At a considerably later stage than the above the infundibulum sends out a pouch-like evagination, which from the beginning is in close contact with the wall of the oral sac and forms the infundibular part of the hypophysis.

Summary and Conclusion.

The foregoing series of sections furnish a clear chain of evidence in favor of the epiblastic origin of the hypophysis in *Chelonia*. Stage B (Figs. 2 and 3) itself is a conclusive proof of such an origin. Here with the pharyngeal membrane yet unbroken we find that the evagination is on the epiblastic side of the membrane. The distinct limiting line which marks the inner border of the hypophysial pouch excludes the supposition that the hypoblast cells may take a part in its formation. Even at the early stage represented in Fig. 1 there is

a distinct limiting line between the epiblast and hypoblast at the point where the hypophysis will later appear.

After the mouth opening has appeared (Stage C, Fig. 4) we find the hypophysis at first in the same relative position as in Stage B. Then the great increase in size of the fore-brain forces the epiblastic pocket to a position far back in the pharyngeal cavity, so that Stages D and E when considered alone would lead one to believe that the hypophysis is of hypoblastic origin in this group. That such is not the case is made evident by following the whole series of changes through all the different stages. There can be no doubt that in the *Chelonia* at least the oral evagination which goes to form the hypophysis is of epiblastic origin. As regards the infundibular portion there is no essential difference of opinion and its development need not be touched on here.

The bearing of the above conclusions on the paleostome theory of Kupffer and the neostome theory of Dohrn is of some interest. According to Kupffer, the hypophysis was originally a canal connecting the fore-gut with the epiblast, and represents an ancestral œsophagus which came up in front of the fore-brain and was replaced by the modern œsophagus at the time when the mouth was forced to a more ventral position by the enlargement of the brain. Dohrn, on the other hand, has picked out the epiphysis and hypophysis as remnants of the old annelid œsophagus which went up through the brain. Both of the above views presuppose some connection of the hypophysis with the hypoblast. The sections of turtle embryos which are described in the present paper give no evidence of such a connection at any stage.

In conclusion I wish to express my sincere thanks to Prof. H. F. Nachtrieb, who suggested to me the investigation of chelonian development and has aided me in many ways during the progress of the work.

DEPARTMENT OF ANIMAL BIOLOGY,
UNIVERSITY OF MINNESOTA, December, 1900.

LITERATURE REFERRED TO IN THE TEXT.

- BALFOUR, F. M. A Monograph on the Development of Elasmobranch Fishes. London. 1878.
- DAVIDOFF, M. VON. Ueber präoralen Darm und die Entwicklung der Prämandibularhöhle bei den Reptilien (*Platydictylus mauritanicus* L. und *Lacerta agilis* Merr.). *Kupffer Festschrift*. pp. 431-454. 1899.
- DEAN, BASHFORD. On the Larval Development of *Amia calva*. *Zool. Jahrb.* 1896.
- DOHRN, A. Studien zur Urgeschichte des Wirbelthierkörpers. *Mittheil. der Zool. Stat. zu Neapel*. Bd. iii. 1882.
- HALLER, B. Untersuchungen über die Hypophyse und die Infundibularorgane. *Morph. Jahrb.* 1897.
- HOFFMANN, C. K. Reptilien, in Bronn's Klassen und Ordnungen des Tierreichs. Bd. vi, Abth. iii. 1888.
- HOFFMANN, C. K. Beiträge zur Entwicklungsgeschichte der Selachii. *Morph. Jahrb.* Bd. xxiv. 1896.
- KUPFFER, C. Studien zur vergleichenden Entwicklungsgeschichte des Kopfes bei Kranioten. Hefte 1-3. München und Leipzig, Lehmann. 1893-95.
- LUNDSBORG, H. Die Entwicklung der Hypophysis und des Saccus vasculosus bei Knochenfischen und Amphibien. *Zool. Jahrb.*, Abth. f. Anat. u. Ontog. Bd. vii. 1894.
- LUSCHKA. Der Hirnanhang und die Stiezdüse. Berlin. 1860.
- MELCHERS, FRITZ. Ueber rudimentäre Hirnanhangsgebilde beim Gecko. *Zeitschrift f. wiss. Zool.* 1899.
- MIHALKOVICS, V. v. Entwicklungsgeschichte des Gehirns. Leipzig. 1877.
- MÜLLER, W. Ueber die Entwicklung und den Bau der Hypophysis und des Processus infundibuli. *Jenaische Zeitschr.* Bd. vi. 1871.
- NUSSBAUM, J. Einige neue Thatsachen zur Entwicklungsgeschichte der Hypophysis cerebri bei Säugethieren. *Anat. Anzeiger*. Bd. xii. 1896.
- ORR, H. Contribution to the Embryology of the Lizard. *Journ. of Morph.* Vol. i. 1887.
- PRATHER, J. M. The Early Stages in the Development of the Hypophysis of *Amia calva*. *Biol. Bull.* Vol. i. 1900.
- REX, HUGO. Ueber das Mesoderm des Vorderkopfes der Ente. *Archiv f. Mikr. Anat.* Bd. i. 1897.
- VALENTI, G. Studio sull' origine e sul significato dell' ipofisi. *Atti d. l'Accad. med.-chir. d. Pavia*. Vol. viii. 1895.

ABBREVIATIONS USED IN CONNECTION WITH THE FIGURES.

<i>Ao.</i>	Aortic arch.	<i>M. B.</i>	Mid brain.
<i>Ca.</i>	Cardiac mesoderm.	<i>Md.</i>	Floor of medullary groove.
<i>Ect.</i>	Epiblast.	<i>Nc. N.</i>	Notochord.
<i>End.₁</i>	"Primary hypoblast."	<i>Pa.</i>	Proamnion.
<i>End.₂</i>	"Secondary hypoblast."	<i>P. G.</i>	Preoral gut.
<i>Ep.</i>	Epiphysis.	<i>P. M.</i>	Pharyngeal membrane.
<i>F. B.</i>	Fore-brain.	. . .	Dotted line above <i>Md.</i> in Fig. 1 represents the height to which the medullary folds have risen.
<i>F. G.</i>	Fore-gut.		Position and direction of the hypophysial evagination.
<i>H. B.</i>	Hind-brain.		Pharyngeal membrane and position of future mouth opening.
<i>H. C.</i>	Canal connecting the two pre- mandibular head cavities.		
<i>Ht.</i>	Heart.		
<i>Hyp.</i>	Hypophysis.		
<i>Hyp. S.</i>	Stalk of hypophysis.		
<i>I.</i>	Infundibulum.		
<i>M.</i>	Mouth opening.		

PLATE I.

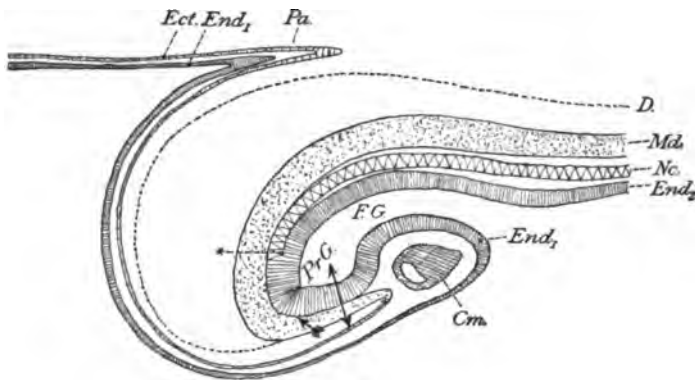


FIG. 1 (Pl. I), Stage A. — Combination diagram formed from the six sections nearest to the median sagittal section. Embryo of *Chrysemys marginata*. Mesoblastic somites = $5\frac{1}{2}$. Lg. of embryo = 2.5 mm. $\times 62$.

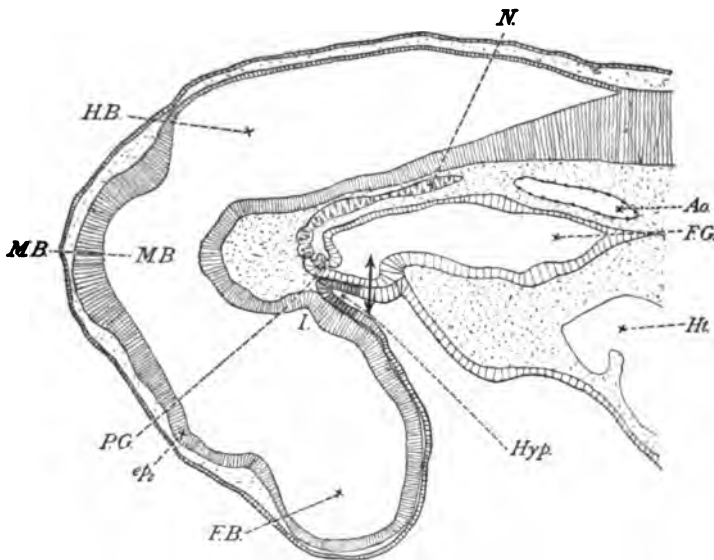


FIG. 2 (Pl. I), Stage B. — Diagram of the median sagittal section of an embryo of *Aspidonectes spinifer*. Lg. = 5.2 mm. Mesoblastic somites = 21. $\times 50$.

PLATE II.

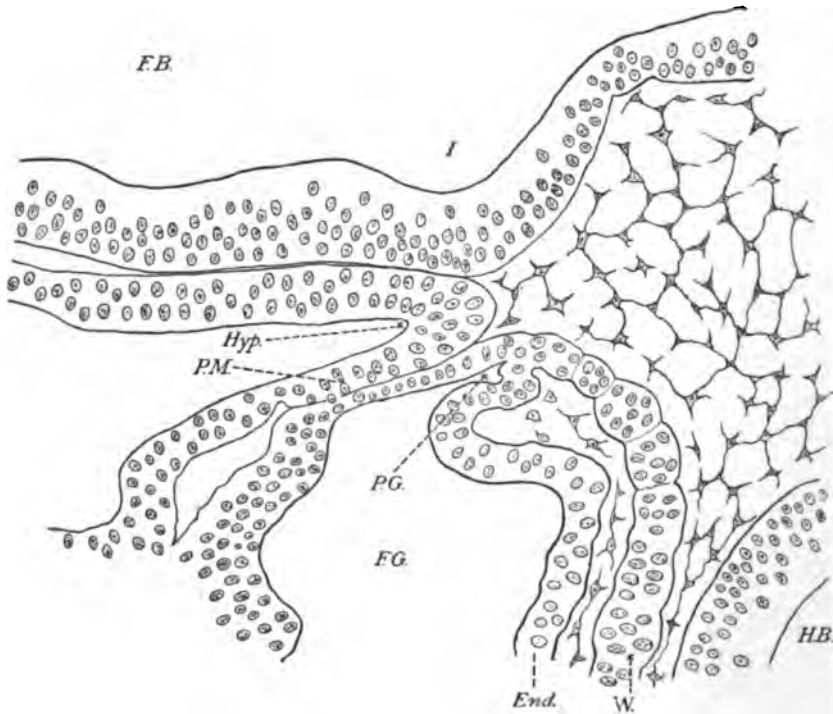


FIG. 3 (Pl. II), Stage B. — Median sagittal section of *Aspidonectes spinifer*.
Part of Fig. 2, but with Magn. $\times 200$.

PLATE III.

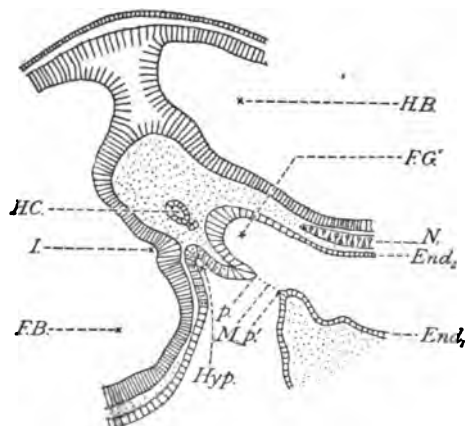


FIG. 4 (Pl. III), Stage C.—Median sagittal section of *A. spinifer*.
Lg. = 5.7 mm. Mesoblastic somites = 21. $\times 50$.

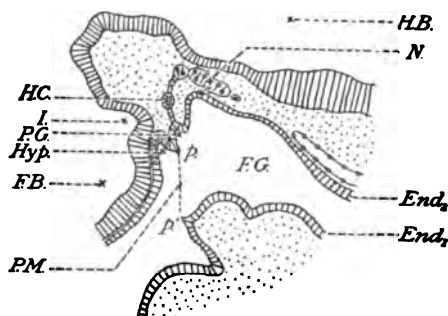


FIG. 5 (Pl. III), Stage D.—Median sagittal section of *A. spinifer*.
Lg. = 6.0 mm. $\times 50$.

PLATE IV.

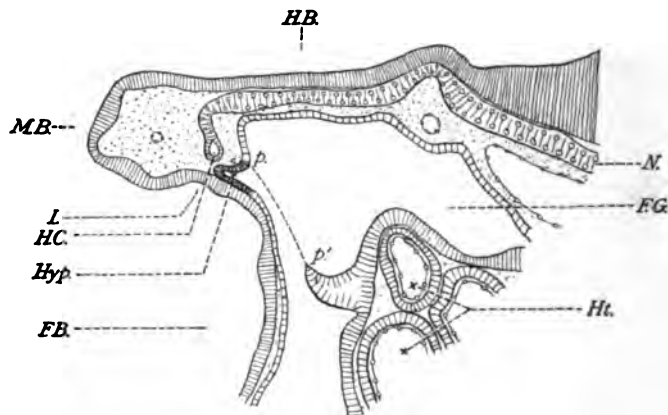


FIG. 6 (Pl. IV), Stage E. — Median sagittal section of *Chelydra serpentina*.
Lg. = 7 mm. × 50.

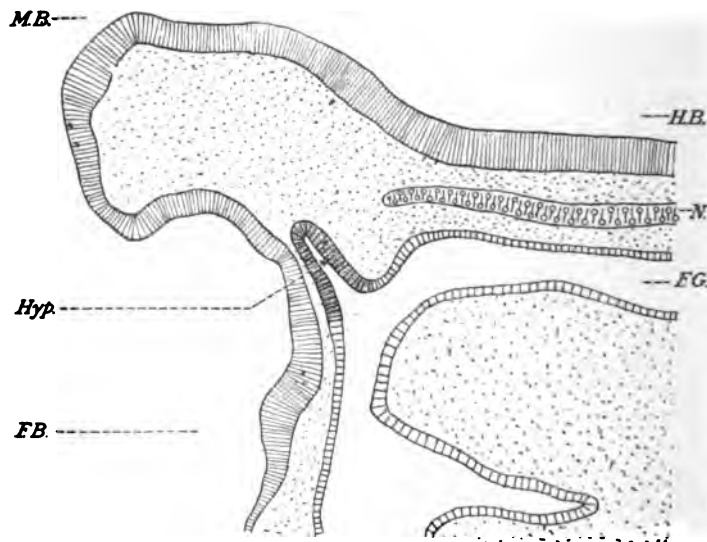


FIG. 7 (Pl. IV), Stage F. — Median sagittal section of *A. spinifer*.
Lg. = 7.5 mm. × 50.

PLATE V.

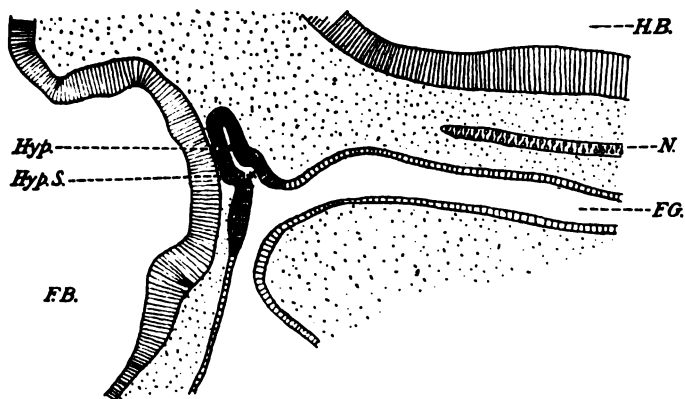


FIG. 8 (Pl. V), Stage F. — Sagittal section from the same series as Fig. 7, but three sections ($40\ \mu$) to the side of the median line. $\times 50$.

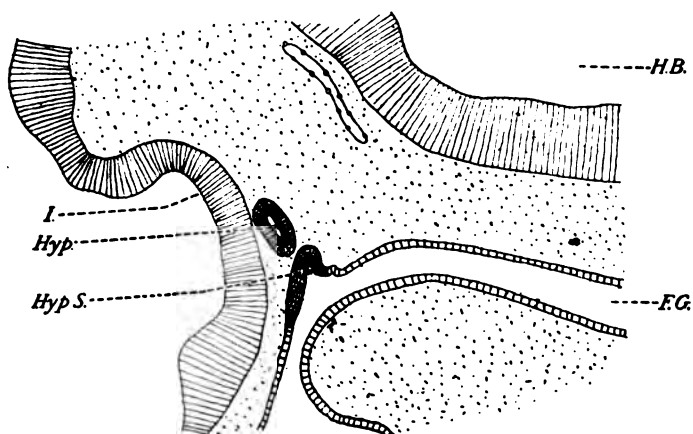


FIG. 9 (Pl. V), Stage F. — Sagittal section from the same series as Fig. 7, but six sections ($80\ \mu$) to the side of the median line. $\times 50$.

ON PHORONIS PACIFICA, SP. NOV.

HARRY BEAL TORREY.

DURING the past summer eight specimens of *Phoronis* came into my hands. Five were collected in June, 1894, in Humboldt Bay, California, by an expedition from the University of California. Three were brought back from Puget Sound by the Columbia University expedition of 1897. As the occurrence of *Phoronis* on the Pacific coast has never been recorded, and it is eminently desirable that all localities in which this interesting form may be obtained should be made known to naturalists, I have undertaken to describe this material, which represents a single species hitherto unknown.

The following table will indicate the distribution and date of first description of all the known species of *Phoronis*. For the species presently to be described I propose the name *P. pacifica*.

<i>P. hippocrepeia</i> Str.	Wright. ¹	1856	Great Britain.
<i>P. ovalis</i>	" "	1856.	
<i>P. (Crepina) gracilis</i>	Van Ben. ²	1858.	
<i>P. Buskii</i>	McIntosh. ³	1881	Philippines.
<i>P. australis</i>	Haswell. ⁴	1882	Port Jackson, N.S.W.
<i>P. Kowalevskii</i>	(Caldwell) ⁵ Benham. ⁶	1883	Naples.
<i>P. psammophila</i>	Cori. ⁷	1889	Messina.
			Port Jackson?
<i>P. Sabatieri</i>	Roule. ⁸	1889	Gulf of Lyons.
<i>P. architecta</i>	Andrews. ⁹	1890	North Carolina.
<i>P. ijimai</i>	Oka. ¹⁰	1897	Japan.
<i>P. pacifica</i> .		1901	Humboldt Bay, California; Puget Sound, Washington.

¹ *Proc. Roy. Phys. Soc. Edin.*, vol. i (1856), p. 165; *Edin. New Phil. Journ.*, vol. iv (1856), p. 313. ² *Ann. Sci. Nat.*, 4th ser., vol. x (1858), pp. 11-23.

³ *Proc. Roy. Soc. Edin.*, vol. xi (1881), p. 211; *Chall. Rep. Zool.*, vol. xxvii (1888), 27 pp. ⁴ *Proc. Linn. Soc. N. S. W.*, vol. vii (1882), pp. 606, 607.

⁵ *Proc. Roy. Soc. Lond.*, vol. xxxiv (1883), pp. 371-383.

⁶ *Quart. Journ. Micr. Sci.*, vol. xxx (July, 1889), pp. 125-158.

⁷ *Dissert. Prag* (Juli, 1889); *Zeitschr. f. wiss. Zool.*, vol. li (1890), pp. 480-568.

⁸ *Compt. Rend. Acad. Sci. Paris*, vol. cix (1889), pp. 195, 196.

⁹ *Ann. Mag. Nat. Hist.*, 6th ser., vol. v (1890), pp. 445-449.

¹⁰ *Annot. Zool. Japon.*, vol. i (1897), pp. 147, 148.

Cori has discussed in an interesting fashion all save the last four species. Roule's *P. Sabatieri* and Andrews' *P. architecta* were apparently unknown to him, though descriptions of both were published before the date of his manuscript. *P. Sabatieri* is known to me only through a meager description. It differs from the other European forms in size and habit, and approaches *P. architecta* in these respects. The latter possesses the simple lophophore and comparatively small number of tentacles (60) of the European species. It may be distinguished from them (with the exception of *P. Sabatieri*, with which it may prove identical) by its larger size, its straight tubes and solitary habit, its strong longitudinal muscles (excepting *P. psammophila*), the presence of a ciliated groove in the digestive tract, and possibly by a separation of the sexes. While it agrees fairly well with *P. Buskii* in size, it differs from that species in the other characters enumerated, as well as in the complexity of the lophophore and the number of tentacles. It is thus more closely allied to the European than to the Australian and Philippine forms.

The differences between *P. australis* and *P. Buskii* are merely of habit and size, which has caused Benham to suggest their identity on the supposition that these differences are due to dissimilar environmental conditions.

The description of the Japanese species has been inaccessible, so that I can state nothing with regard to it save its existence.

It is an interesting fact that no one has cared to segregate the species of *Phoronis* under more than one generic name, and indicates the trifling character of the differences which serve to distinguish them. We may separate them, however, into two groups widely separated geographically. In the one belong the European forms, including, perhaps, the American *P. architecta*. In the other belong *P. australis* and *P. Buskii*. We may disregard the Japanese species on account of dearth of information, and the *P. psammophila* which Haswell has found at Port Jackson and which may have been brought from the Mediterranean on a ship's bottom.

P. pacifica occupies a place intermediate between these two groups both geographically and anatomically, but is somewhat

more closely related to the American than to the Australian species. In size it resembles *P. Buskii*, as well as in the complexity of the lophophore; though instead of three coils in the

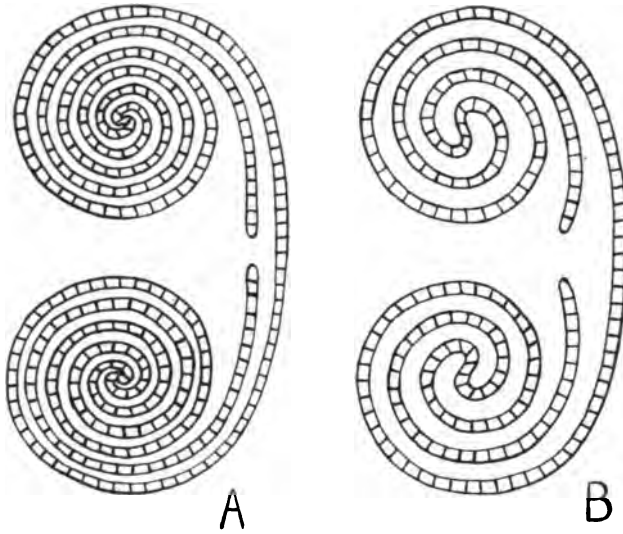


FIG. 1. — Diagrammatic cross-sections of the lophophores of *P. australis* or *P. Buskii* (A), *P. pacifica* (B).

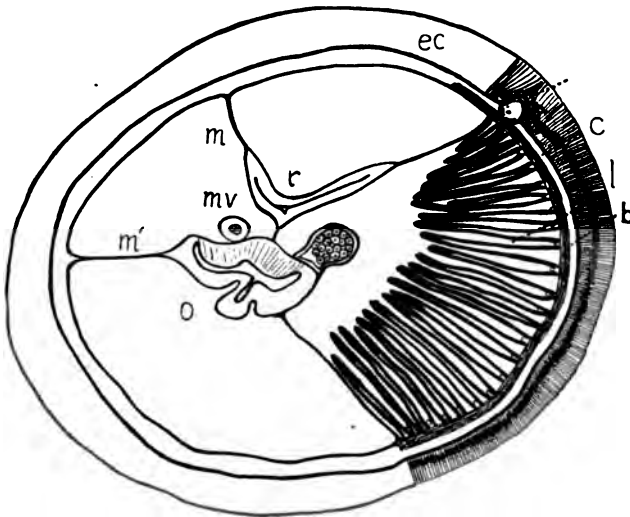


FIG. 2. — Section through the upper third of *P. pacifica*; but one quadrant detailed; semi-diagrammatic. *b*, basement membrane; *c*, circular muscle; *ec*, ectoderm; *l*, longitudinal muscles; *m*, *m'*, mesenteries; *mv*, median vessel; *n*, nerve; *o*, oesophagus; *r*, rectum.

spirals of the lophophore there are one and one-half to two, and a correspondingly smaller number of tentacles (170-200),



FIG. 3. — Cross-section of one longitudinal muscle ridge of *P. pacifica*, slightly broken.

Fig. 1, *A*, *B*. In the strength of its longitudinal muscles (Figs. 2, 3) it departs from *P. Buskii* and even surpasses *P. architecta*, resembling the latter species in the possession of a specialized ridge in the digestive tract (though this does not pass into the groove that Andrews describes), in the structure of the nervous system, lophophore organ and tube, in habit, and in the possible separation of the sexes.

It is not my intention to enter into a full and detailed anatomical description of *Phoronis*, which the labors of Benham and Cori have rendered largely unnecessary. But a few words on some points may not be out of place.

In one of the Puget Sound specimens spermatogonia and spermatocytes were found packed around the blood vessels in the aboral regions of the body, but no spermatozoa nor ova. The aboral ends of all the Humboldt specimens were wanting, so it was impossible to determine whether they were monoecious or dioecious. In one nephridium three ova were found unaccompanied by spermatozoa; the first polar body was forming in one. As there was no sign of spermatozoa in these eggs, it is probable that in this species fertilization takes place either externally or within the nephridium. It is quite possible that the sexes may be separate, or ova and sperm may be produced by the same individual but not simultaneously.

The blood corpuscles have a conspicuous yellow color and measure 10-15 μ in diameter.



FIG. 4. — Cross-section of the oesophagus of *P. pacifica*, showing the ridge, its base in contact with the median longitudinal blood vessel.

The ciliated ridge was present for a considerable distance in the oesophagus, but could not be seen in the stomach either as a ridge or a groove. In Figs. 2, 4 it is indicated in section, where it appears to be a shallow groove, an appearance probably due to the folding of the wall of the oesophagus. Its position relative to the longitudinal blood vessels is identical with that described for *P. architecta*. The nuclei stain more intensely with haematoxylin than the other nuclei of the oesophagus, and are crowded together usually in several layers. These facts make the area quite conspicuous in section.

The muscles reach their greatest development in the oral third of the body, where they form more than eighty high narrow ridges. In the aboral third these are reduced to a very inconspicuous layer, though still retaining their identity, being separated throughout their length by characteristic folds of peritoneal epithelium.

There is a delicate peritoneum covering the muscle ridges, the nuclei only (Fig. 3, *n*) being seen with ordinary powers of magnification. Occasionally a similar nucleus is found within the fold of muscle (*n'*).

The nervous system is constructed as in *P. architecta*, with one interesting exception. The two longitudinal cords, which are of exceedingly unequal length, instead of ending in the nerve ring of the lophophore, are continuous across the median line at the level of the median mass of ganglion cells. The loop thus formed is closely applied posteriorly to the latter and touches the lophophore nerve on each side of the rectum, apparently without fusing at either point. Just how intimate this contact is cannot be determined from my poorly fixed material. The brevity of the descriptions of this portion of the nervous system in *P. architecta* and other species leads me to suspect that the seemingly exceptional condition in *P. pacifica* may prove to be of more general occurrence.



FIG. 5.—Semi-diagrammatic cross-section of lophophore organ within the concavity of the lophophore.

The lophophore organ is extremely variable and may be present or absent, as in *P. psammophila*. It may resemble that of *P. australis*, though differing somewhat in shape (Fig. 5). In this case it is simple, with a thickened glandular epithelium lying for the most part against the inner circle of tentacles, and an outer free non-glandular edge of much lower cells. In another case, however, it had the form of the same organ in *P. architecta* and *P. psammophila*, as described and figured by Andrews and Cori, being composed of a basal lobe and a distal "carpel-like organ." This condition seems to have been attained by the addition of the "carpel-like organ" to the structure (basal lobe) which corresponds to the entire organ in *P. australis*.

The following is a diagnosis of the species, from material preserved in alcohol and formalin :

Total length may be 9 cm., of which the tentacles represent from $2\frac{1}{2}$ to 4 mm.

Diameter, $1\frac{1}{2}$ to 2 mm.

Lophophore spirally coiled, each spiral possessing from $1\frac{1}{2}$ to 2 complete turns.

Tentacles 170 to 200.

Lophophore organ present or absent; extremely variable in form.

Each animal occurs singly and completely fills tube.

Tube straight, cylindrical, composed of delicate chitin, encrusted with fine sand grains.

Ridge of thickened epithelium in the descending limb of the digestive canal, just beneath the median longitudinal blood vessel.

Longitudinal nerve trunks unite across median line between mouth and anus.

Longitudinal muscles in numerous very high and narrow folds which reach their maximum in the distal third of the body.

Sexes possibly separate.

Localities: Puget Sound, Washington; Humboldt Bay, California, on sand and mud flats that may be uncovered by the tide.

COLUMBIA UNIVERSITY, January, 1901.

ON MUSCLE REGENERATION IN THE LIMBS OF PLETHEDON.

ELIZABETH W. TOWLE.

SPALLANZANI (1768) and Bonnet (1777) showed that a salamander whose limbs have been cut off has the power to regenerate new ones. This discovery has been confirmed by later writers, and although some histological work has been done, yet the method of regeneration of the muscle bundles has not been worked out. There are several possibilities: first, the old fibers might break down at the cut ends and the new ones develop from the indifferent tissue so formed, each old muscle thus completing itself independently. Or, the cut muscles might degenerate along their entire length, and new ones take their place; or some of the old muscles might degenerate, new ones being formed from this tissue, while some fibers might break up into smaller new fibers. An attempt has been made in this work, not so much to follow the origin of the cells in detail as to discover the general processes taking place in the leg that lead to the formation of the new muscles. The regenerating limbs of *Plethodon cinereus* were used. They were studied by means of serial sections.

In addition to this histological study, I have also experimented on a number of American urodeles in order to see in which ones regeneration of the limbs takes place. For this purpose a number of the commoner forms have been studied, and in connection with these results a statement is given of the previous observations on European forms.

I.

Method.—One of the anterior limbs of *Plethodon cinereus* was removed halfway between elbow and hand. The regenerating limbs were put up at intervals varying from four days

to two weeks. They were fixed in corrosive acetic, hardened for two or three days in 95% alcohol, and then decalcified for from six to eight days in a nitric acid solution (HNO_3 sp. gr. 1.42, 2 vols. + H_2O , 98 vols.) which was changed daily. They were finally hardened again for three days in 95% alcohol, embedded and cut. Some limbs were stained *in toto* with borax carmine, but the best results were obtained by the method used by Byrnes ('82), *viz.*, staining on the slide in Delafield's haematoxylin, followed by a wash of picric acid in absolute alcohol. This latter method differentiates the muscle substance very clearly.

Eleven stages were preserved at the following intervals :

Time of Operation.	Time of Killing.	Age of Stump.
1. May 4, 1900. ¹	May 14, 1900.	7 days.
2. " " "	" 18, "	11 "
3. " " "	" 29, "	22 "
4. Oct. 22, 1899.	Nov. 20, 1899.	29 "
5. " " "	Dec. 5, "	44 "
6. " " "	" 15, "	54 "
7. " " "	" 15, "	54 "
8. " " "	Jan. 5, "	75 "
9. " " "	" 5, "	75 "
10. " " "	" 20, "	90 "
11. " " "	March 22, "	151 "

Transverse sections of this series were cut. Nos. 1, 2, and 3 were stained with haematoxylin and picric acid ; 4, 5, and 6 with borax carmine ; 7 with borax carmine and picric acid ; 8 and 9 with Biondi-Ehrlich solution ;² 10 and 11 with haematoxylin and picric acid. In addition, normal limbs were cut and stained in a similar manner and used for comparison.

Results. — For convenience in description I shall consider the sectioned limb as made up of three Regions : I, that between the cut and the elbow ; II, the Region just above the cut ; and III, the growing end. In the earliest stages Region III does not, of course, exist.

¹ It will be noticed that stages 1, 2, and 3 were preserved later in the year, but observations will be described in the above order.

² This stain was not successful, and the stages were replaced by one stained with haematoxylin and picric acid.

In the first of the series of transverse sections changes in the cut muscles are already noticeable, the most striking being the increase in the number of nuclei, especially in the outer fibers of the limb. This increase can be seen in Region I as far up as the origin of the muscles at the elbow. In the outer fibers the muscle tissue is becoming thinner and disappearing, and while the outlines of fibers and bundles are not lost, they are much less clear than in the normal limb. The inner bundles, however, are but little affected, and extend unbroken to the cut end, which is at this time not yet entirely covered by ectoderm. No mitosis is seen in this section.

In stage 2 the changes are more marked. There has been a continued increase in the number of nuclei in Regions I and II, the outlines of the outer fibers and bundles are lost, while the muscle substance has disappeared except for disintegrating fragments here and there, contrasting sharply with the thin cytoplasm of the neighboring cells. The inner fibers still extend to the end of the limb, which is now covered entirely by several layers of ectoderm. In the neighborhood of the cut three or four mitotic figures are to be found. In the third stage Region III begins to appear as a small knob of undifferentiated tissue behind the cap of ectoderm. In this knob and for a short distance above it among the outer cells karyokinesis is not uncommon.

If we compare the following stage (4) with the normal limb, the principal changes that have taken place will be very clearly brought out. In Region I the increase in number of nuclei is very great. Even as far as the elbow two to four nuclei may be found in a section of a single fiber, often crowded together so as almost to fill it (Fig. 1). Many nuclei are also scattered between the fibers. Below the elbow the number of nuclei increases, the outlines of the outer fibers are completely lost, and the outer half of the limb, which is normally solid muscle, is seen to be made up of a dense mass of nuclei surrounded by loose protoplasmic substance, with here and there



FIG. 1.

clumps of disintegrating muscle tissue. The outlines of the inner fibers are somewhat less distinct than in the preceding stage, and some of the bundles seem to be splitting up. This condition is represented in Fig. 2. Most significant is the fact that at no stage is any karyokinesis found among the muscle fibers, although the increase in the number of nuclei is enormous. Passing through Region II of this stage, the number of old fibers decreases and the scattered nuclei increase, until at the



FIG. 2.

plane where the cut was made all old fibers disappear and we reach Region III, which is made up entirely of closely crowded nuclei, each surrounded by a small amount of protoplasm. Karyokinesis is first seen in II, a short distance above the cut, among the outer cells, *never in the muscle fibers*, and the number of dividing cells increases toward the growing tip until it becomes quite large.

In the fifth stage a further difference is to be noted. We find in Region I, in the inner part of the limb, the old muscle

fibers, often with several nuclei, and broken frequently into quite small fragments. Outside these are numerous nuclei, as before, but now surrounded by very distinct muscle tissue (Fig. 3). This tissue has formed often about several nuclei in a group, and has no distinct walls; there seem to be as yet no distinct muscle fibers. But the line of separation between the old muscle fibers and the new tissue is distinct, and a few old fibers can be traced to the region of the cut, although at that level the greatest part of the tissue is new. New muscle substance has appeared about the nuclei for a short distance below the cut; but it decreases as we pass down, until we find only crowded nuclei and thin protoplasm. In this region, as before, numerous karyokineses are seen.



FIG. 3.

In Region I of the sixth stage all the muscle fibers are small and the definite line between the old and new is lost. The majority of the fibers contain in cross-section but one nucleus, though some may contain two or three, and in general the smallest fibers and most nuclei are on the outer side of the limb. This is especially noticeable in Region II, where the outer (new) fibers are exceedingly small. The muscle tissue decreases in amount as we pass to Region III, until it is all lost. Cells dividing by karyokinesis appear at this level.

Stage 7, though of the same age as the preceding, is somewhat further differentiated, and in this the new fibers are more rounded and have assumed a more characteristic form (Fig. 4). A comparison with the normal shows smaller fibers

and great excess of nuclei, there being often two or three to one fiber and many outside the fibers.

The later stages need not be described in detail. As the limb grows longer the formation of new muscle tissue progresses farther down toward the tip, the new fibers being always small and containing several nuclei. The number of nuclei outside the fibers decreases, until in stage 11 the muscles look quite normal, and the number of nuclei is excessive only in the region of the foot, which is at this time

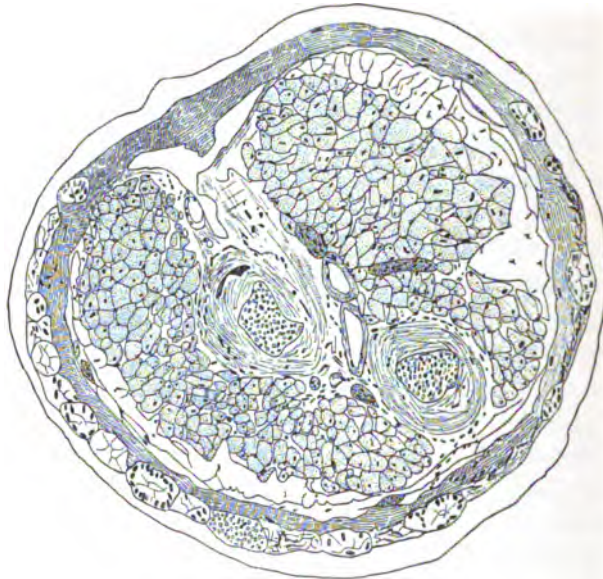


FIG. 4.

clearly differentiated. Karyokinesis is found, I believe, without exception, near the growing end, never in the upper regions.

The first appearance of any definite grouping of cells appears in stage 5, where the arrangement into bundles is foreshadowed. As the fibers form, the division into bundles becomes more distinct, until in stage 11 they are all differentiated as far down as the foot, and here we can see by the arrangement of nuclei where the bundles are to be.

In the process of regeneration described above there are certain things to which I wish to direct especial attention. In

the first place there is a great increase in the number of nuclei *within the old fibers*, but in no case is any karyokinesis found there. This degenerative process in the old fibers must therefore take place by direct division of the nuclei. Instances of this division are shown in Figs. 5 and 6. To this division and to the disintegration of some of the old fibers is due the enormous accumulation of nuclei in the outer part of the limb (Fig. 2). The cells so formed then begin to divide by karyokinesis in the region of the cut, and thus a further increase in their number takes place. In these outer cells new muscle tissue forms and the new fibers are built up. A certain



FIG. 5.

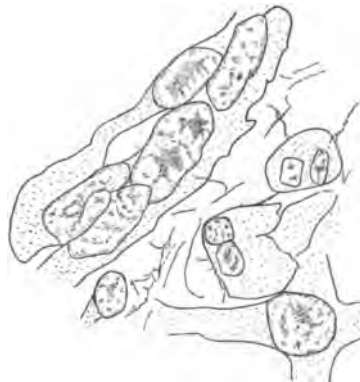


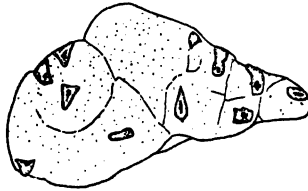
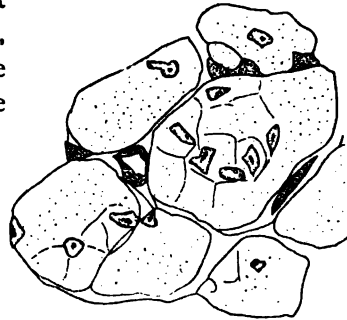
FIG. 6.

number of the old fibers remain in the middle of the limb, and in these the muscle tissue never disintegrates, though it splits longitudinally.

Again, as at any one level the number of nuclei far exceeds the number of fibers in a normal muscle, a great number of them must, between the early stages and the fully formed limb, either degenerate or be transported (*cf.* Figs. 3 and 4). That this is so is easily seen, for two reasons: (1) when the new fibers form, at a given level several nuclei are often included in one fiber; when the limb is full-grown there is only one; (2) among the newly formed fibers, but *between* them, are many scattered nuclei; the majority of these disappear in later stages.

One further point should be mentioned. In stage 5 the line of distinction between new and old fibers is clear, owing to their difference in size. In stage 7 this distinction has disappeared. This is due not only to an increase in the size of the new fibers, but to a decrease in the old. This decrease is, I believe, due to the longitudinal splitting of such of the old fibers as are left (Fig. 7. *a* and *b*).

Beside the stages described above, four others were preserved and cut longitudinally. This was a somewhat difficult operation, for the new part forms at an angle with the upper arm, and it is hard to orient the piece in such a way as to insure

FIG. 7 *a*.FIG. 7 *b*.

true longitudinal cutting of the muscle fibers. The material was preserved at the following intervals:

Time of Operation.	Time of Killing.	Age of Stump.
1. Jan. 29, 1900.	Feb. 2, 1900.	4 days.
2. " " "	" 12, "	14 "
3. " " "	" 19, "	21 "
4. " " "	" 28, "	30 "

The first of these shows no transformation at the cut ends of the muscles, but the ectoderm has closed in over the wound. In the second there is a considerable increase in the thickness of the ectoderm, and under it a collection of scattered nuclei, exactly similar to the tissue in the growing end of later stages. As the limb grows in length this tissue increases in amount and a number of mitoses are seen in it. Degeneration of the old fibers is distinctly noticeable in stage 3, and fibers are found, as before, filled with nuclei, but there is no karyokinesis in any muscle fiber.

SUMMARY.

The main changes that take place in the muscles of *Plethodon cinereus* during the process of regeneration are as follows :

1 (a) In the cut muscles the nuclei divide directly and in the outer bundles the fibers disintegrate, leaving masses of nuclei with a small amount of cytoplasm. (b) Some of the cells so formed later divide mitotically, and by them new muscle substance is laid down. (c.) As the number of nuclei is, however, far in excess of the normal number of muscle fibers, many nuclei must degenerate or be transported.

2. The fibers of the inner bundles do not disintegrate, but split longitudinally, giving rise to smaller fibers, which are soon indistinguishable from those formed as described in 1.

3. The arrangement into muscle bundles first becomes clear at the end of about six weeks.

4. There is no change in the muscles of the upper arm.

II.

The following is a brief summary of the main observations that have been made on the power of regeneration of the limbs in European species of salamanders.

Spallanzani (8), in 1768, published a number of observations on aquatic salamanders, presumably species of *Triton*. In these he found that any or all of the limbs will regenerate, no matter what the species, size, or age of the animal, the larger ones regenerating more slowly than the smaller forms. Regeneration will take place, he maintained, even when the limbs are disarticulated.

Bonnet, 1777 (2), confirmed Spallanzani's observations, finding that in *Triton cristatus* the hands and fingers will regenerate.

Von Siebold (7), in 1828, recorded abnormal regeneration of the fingers of *Triton cristatus*.

Higginbottom, in 1847 (4, p. 29), observed that in *Triton* the limbs will regenerate at a temperature of from 48° to 57° F.

Philippeaux (6), in 1866, was the first to prove conclusively that limbs when disarticulated will not regenerate. His experiments were made on *Triton cristatus* and *Axolotl*.

Dumeril (3), in his paper of 1867, in the course of other observations, notes the fact that in an Axolotl the two anterior limbs regenerated after injury.

Wiedersheim, 1875 (1, p. 95), found that the toes of *Triton cristatus* will regenerate, while there is no regeneration of the limbs in *Proteus* and *Siren lacertina*.

Erber, 1876 (4, p. 34), notes regeneration of the feet of *Siren lacertina*.

Goette, 1879 (5), records the regeneration of a leg of *Proteus* after a year and a half. Regeneration also occurs in *Amphiuma* and *Siren*, in *Triton cristatus*, *T. taeniatus*, and their larvae.

Weismann (9), in *The Germ Plasm*, 1893, says that the limbs of *Salamandra* regenerate, while in *Triton marmoratus* regeneration is slight or absent.

Barfurth (1), in 1894, reports regeneration of the feet and digits of *Triton taeniatus* and *Siredon pisciformis*. This regeneration is normal or abnormal according to the plane and method of the injury.

I have experimented on the following forms, removing a fore foot and a hind foot from different individuals of each species: *Plethodon cinereus*, *Spelerpes ruber*, *S. guttolineatus*, *Desmognathus ochrophaea*, *Manculus quadridigitatus*, *Amblystoma opacum*, *Diemyctylus viridescens*, *Amphiuma means*, and *Necturus maculatus*. Of these, all have regenerated.¹ The regeneration in *Spelerpes*, *Desmognathus*, *Manculus*, and *Amblystoma* was comparatively rapid, and new limbs were well formed in four months, though they were somewhat smaller than the old limbs. *Diemyctylus* was slower in reaction, while the first *Necturus* to show a distinct regenerated stump did so only after eight months. Other individuals of the same species showed no regeneration even at that time.

I desire to thank Professor T. H. Morgan, under whose direction this work was undertaken, for his kindly assistance during its progress.

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¹ *Amphiuma means* was observed for only eleven weeks. At that time regeneration was slight. The regenerated stump was not sectioned.

LITERATURE.

1. BARFURTH. Arch. f. Entwicklungsmechanik der Organismen. 1895. p. 91.
2. BONNET. Mémoire sur la reproduction des membres de la salamandre aquatique. *Œuvres complètes*. Tome xi. I. Mémoire. 1777.
3. DUMERIL. Nouvelles archives du muséum d'histoire naturelle de Paris. Tome iii. 1867. p. 119.
4. FRAISSE. Die Regeneration von Geweben und Organen bei den Wirbelthieren. Cassel und Berlin. 1885.
5. GOETTE. Ueber Entwicklung und Regeneration des Gliedmassenskeletts der Molche. Leipzig. 1879.
6. PHILIPPEAUX. Comp. Rend. 1866. p. 576.
7. SIEBOLD, VON. Observationes quaedam de salamandris et tritonibus. *Dissert. Berolini*. 1828.
8. SPALLANZANI. Précis sur les reproductions animales. 1768. p. 79.
9. WEISMANN. The Germ Plasm. New York, Charles Scribner. 1893.

THE FACTORS THAT DETERMINE REGENERATION IN ANTENNULARIA.

T. H. MORGAN.

THE following experiments were carried out at the Naples Zoölogical Station during June and July, 1900. As there may be no opportunity in the immediate future of completing the observations, I have determined to publish them as they stand, in the hope that the results may stimulate some one, so situated as to obtain the necessary material, to take up the questions here raised and to bring them to a more satisfactory conclusion.

Loeb's experiments on *Antennularia*, made in 1892, show that pieces of the stem suspended in sea water always regenerate roots at the lower end and a new stem at the upper end. The result was the same whether the apical or the basal end of the piece was uppermost, *i.e.*, whether the piece had a normal or a reversed orientation. Similar results were obtained when pieces were suspended obliquely, the high end producing always the new stem and the low the new roots, etc. These results are similar to certain results that have been obtained in plants, although Vöchting has shown conclusively in many forms that the polarity of the piece is a much stronger factor in determining the regeneration than is gravity. Loeb drew the natural inference from his results, *viz.*, that gravity determines the kind of regeneration that takes place at the ends of the piece. Driesch,¹ who examined later the regeneration of *Antennularia*, found that when a piece of the stem is so placed "that its basal end is freely surrounded by water," a large number of roots are formed from that end. If the end with its roots is cut off, there is generally formed from the cut end a few new roots, but also always a more or less delicate stem composed of a few tubes. This stem is negatively geotropic.

¹ Driesch, H. Studien über das Regulationsvermögen der Organismen, I. *Roux's Archiv.* Bd. v, p. 383.

If the same end is again cut off, there develops rarely one or a few roots, but generally two or three vigorous stems. If the operation is repeated a fourth time, one or two stems are without exception produced.

There is no statement made by Driesch as to how these pieces were orientated with regard to gravity, but the results show that another factor than gravity has an influence on the regeneration. Unfortunately nothing is said with regard to what has taken place at the other end of the piece. I shall try to show that it is not improbable that this may be also a factor in the result, and if so it is possible that Driesch's results are due to this rather than to the action of the water on the free basal end, or at least both factors may be present.

My experiments were primarily undertaken in order to see how pieces would behave when fixed to a revolving wheel, but on account of the apparent disagreement between Loeb's and Driesch's results, it was first necessary to repeat the experiment of suspending pieces with two cut ends in order to see how far gravity acted upon them. In one series of experiments pieces were suspended in an aquarium by means of a silk thread. Some of these had the apical end upwards, others the basal end upwards, and still others were suspended horizontally. In nearly all cases roots developed in the course of a few days from both ends. If the ends were cut off, new roots developed again on both ends; although in one or two cases in which the apical end was uppermost a stem developed at that end. The pieces were from 3 to 5 cm. long.

By means of another device the experiment can be much more satisfactorily carried out. A small square piece was cut from a sheet of cork and a hole bored in its middle. The end of a glass rod, about 20 cm. long or longer, was pushed through the hole in the cork. If the piece of cork is neither too large nor too small, the glass rod, when put into an aquarium, will sink to the bottom until one end touches, but the other end will be held up in a vertical position owing to the buoyancy of the cork.

Pieces of the stem of *Antennularia* were fastened to the sides of the cork by means of two dried cactus spines, that were crossed over the stem and stuck into the cork.

In these experiments, made with pieces of different lengths, and from different parts of the old stem, the results were the same as before. In nearly every case roots developed from both ends, and even after these ends had been once removed. The experiments extended over two or three weeks. Whether, if continued longer, a stem would develop at the upper end among the roots there present, I do not know, but the results suffice to show that the most characteristic thing that occurs is the production of roots from both ends.

I was, therefore, not a little surprised to find in another series of experiments that a different result occurred. I placed in an aquarium a number of pieces of *Antennularia* that remained attached to the stones on which they had been found growing. Most of the pieces stood up vertically from the floor of the aquarium with the apical end upwards; a few pieces I suspended in an inverted position, *i.e.*, with the apical end downwards and the attached basal end upwards. In the former cases the apical ends did not produce roots at all, but a new stem. In the latter cases, in which the pieces were inverted, the apical end produced neither roots nor stem. Although these pieces were observed for only ten days, the time is ample to show that the pieces behave differently from pieces with two cut ends. I regret that I could not carry these experiments further.

One other result should be described, since it seems to have a direct bearing on the last experiment. In one case a very small piece had sunk to the bottom of a dish of water, where it stood with its basal end in contact with the glass. It lay there undisturbed, and attached itself at its basal end by means of new roots. *The apical end produced a shoot.* This result, taken in connection with the preceding experiment, seems to indicate that the development, or the presence of roots on the basal end, prevents the development of roots on the apical end. This result, if it prove constant, opens the way for several interesting experiments that so obviously suggest themselves as to require no further mention.

A few experiments were made with a rotating wheel constructed for the purpose. The wheel consisted of two parallel

rings of wire between which, at equidistant points, were sixteen paddles (5 cm. $\times$ 8 cm.) made of oblong pieces of sheet-copper; spokes (13 cm. long) attached the rings to an axis that rotated in two sockets. When the wheel was immersed in the water of an aquarium, and a stream of water from the tap was made to play (beneath the water) on the plates, the wheel slowly revolved, making about five and one-half revolutions in a minute.¹

Pieces of *Antennularia* were attached to the wheel in the following way. Sheets of cork of the same size as the copper plates were attached to the underside of the latter by wire or string. Pieces of the hydroid were fixed to the cork in different positions by means of crossed cactus spines. The pieces were, on an average, about 15 cm. from the axis of rotation. The results were entirely negative. None of the pieces produced either roots or stems; and the pieces died sooner than did those in other experiments. As this experiment was carried out in a different aquarium, I cannot be certain that the death of the pieces was not due to other causes than to the rotation. Furthermore, it is not evident from the experiment whether the rubbing of the moving ends against the water suppressed the regeneration, or whether the result is due to the continuous change of position in regard to gravity. The rotation was too slow for the action of the centrifugal force to have played any important part. Since other experiments have shown that roots may develop at both ends of a piece suspended vertically, it is improbable that in the rotating pieces the changing position in regard to the action of gravity can account for the result, and it is much more probable that the motion of the piece through the water interfered with the regeneration at the ends. The experiment needs to be repeated more often, and other check experiments carried out in the same tank.

The work that I have done on the regeneration of *Antennularia*, while incomplete in many ways, shows at least that other factors than gravity enter into the result. I do not question the main part of Loeb's results, for they seem to show that gravity

¹ This wheel was left at the Naples Station in the hope that it might be used by some one to continue the experiments.

is a factor in the regeneration of this form ; but the development of roots at both ends that first takes place, as I have found, but which Loeb did not observe, and the behavior of pieces attached at one end, as described in the preceding pages, show that the factors determining regeneration are more involved than previous results seem to indicate.

BRYN MAWR COLLEGE, Feb. 4, 1901.

MENDEL'S LAW OF DICHOTOMY IN HYBRIDS.

C. B. DAVENPORT.

IN the study of hybrids we must, as De Vries (1900b) truly says, no longer pay primary attention to the degree of difference between the forms united — to whether they are species, subspecies, or varieties — but to the behavior of the peculiar characters by which the crossed individuals and their ancestors are distinguishable. For each of these somatic characters corresponds to some peculiarity of the germplasm. The behavior of the differing characters when united in the hybrid is diverse; three categories have long been recognized (Galton, 1888, p. 12). These are: (1) *blending* heritage, illustrated by skin color in man; (2) *alternative* heritage, illustrated by human eye color; and (3) *mixed* heritage, illustrated by the piebald condition of the progeny of mice of different colors. The law of dichotomy in hybrids applies only to the second class, — alternative heritage, — although it has recently been brought forward by De Vries (1900) as the almost universal law of inheritance in hybrids. The law itself was first enunciated very clearly and completely by Mendel (1865) and deserves to bear his name. The law was, however, forgotten. It has been rediscovered independently by De Vries and by Correns (1900), both of whom are able to add new evidence of its validity (for *alternative* heritage!).

In his illustration of Mendel's law, De Vries first classifies hybrids into monohybrids, dihybrids, and polyhybrids, according as their parents differed in one character only, or in two characters, or in many characters. The case of inheritance in monohybrids is the simplest, and will be first considered. Mendel's and De Vries's investigations have established the following principles:

1. Of the two antagonistic peculiarities the hybrid exhibits only one; and it exhibits it completely, so as not to be

distinguishable in this regard from one of the parents. Intermediate conditions do not occur [in *alternative* heritage].

2. In the formation of the pollen and the egg cell the two antagonistic peculiarities are segregated; so that each ripe germ cell carries either one of these peculiarities.

Of the two antagonistic peculiarities united in the hybrid, that which becomes visible in the soma is called by Mendel the *dominating*; that which lies latent is called the *recessive* character. What determines which character shall be dominating is still unknown, and the determination of this point offers an enticing field of inquiry. In some cases the dominating form is the systematically *higher*; in others it is the older or ancestral form.

The law of dichotomy may now be developed. When a hybrid (monohybrid) fertilization takes place the zygote contains both the dominant quality (abbreviated *d*) and the recessive quality (*r*). In the early cleavages *d* and *r* are both passed over into both the daughter-cells; but apparently, at the time of segregation of the germ cells, the somatic cells are provided with *d* only, while the germ cells retain both qualities. In the ripening of these germ cells, probably in the reduction division, *d* and *r* come to reside in distinct cells, so that we have

of the female cells 50% *d* + 50% *r*, and
of the male cells 50% *d* + 50% *r*.

If now hybrids are crossed haphazard, a male *d* cell may unite with either a female *d* cell or with a female *r* cell; likewise a male *r* cell may unite with a female *d* or a female *r* cell. Consequently in the long run we shall have of all the zygotes

25% *d, d* + 50% *d, r* + 25% *r, r*,

or 50% of the zygotes hybrid and 50% of pure blood, and of the latter half exclusively maternal and half paternal. But since the soma developed from the hybrid germ cell has the dominant character, we shall have

75% of the cases with the dominant character
25% " " " " " recessive "

and this agrees with various empirical results, of which the following from Correns is instructive. A cross was obtained

between a species of pea with a green (*g*) germ and one having a yellow (*y*) germ. Yellow is dominating.

Gen. 1.	31 <i>y</i> (hybrid) peas; produced 12 plants; these bore:		
	775 <i>y</i> (hybrid + <i>y</i>) peas (= 75.8%). 21 plants were produced.		247 <i>g</i> (pure-blooded) peas (= 24.2%).
Gen. 2.	7 (33%) pure-blooded <i>y</i> , because they bore:	14 (66%) hybrids, because they bore:	20 plants bore:
	292 <i>y</i> peas.	462 <i>y</i> (hybrid + <i>y</i>) peas (= 76.4%).	149 <i>g</i> (pure-blooded) peas (= 23.6%).
Gen. 3.			670 green peas.

It is clear that if this process of crossing of the hybrids continues, the *proportion* of hybrids to the whole population will diminish; for the share of pure-blooded forms breeds true; while the originally equal share of hybrids is repeatedly halved.

If the hybrid is crossed with one of the parents instead of with another hybrid, we will

$$\begin{aligned} \text{get } (d + r) \ d = d, d + d, r, \text{ and} \\ (d + r) \ r = d, r + r, r. \end{aligned}$$

In the first case all of the progeny will appear of the dominant type. In the second case one-half will appear of that type. This again agrees with experiment.

In the case of dihybrids the law of alternative heritage is somewhat more complicated. Imagine a lot of ripe germ cells with the antagonistic qualities of any pair separated according to the second principle stated at the outset. *A* indicates the one pair of qualities and *B* the other; then we shall have nine classes of zygotes, the proportion of each of which is as follows:

$$\begin{aligned} A. & \quad 25\% \ d, d & \quad 50\% \ d, r \\ B. & \quad 6.25\% \ d', d'; \ 12.5\% \ d, r; \ 6.25\% \ r, r. & \quad 12.5\% \ d, d; \ 25\% \ d, r; \ 12.5\% \ r, r. \\ & \quad A. & \quad 25\% \ r, r \\ & \quad B. & \quad 6.25\% \ d, d; \ 12.5\% \ d, r; \ 6.25\% \ r, r. \end{aligned}$$

Thus the first class has 6.25% purely dominant in both characters; the second class, 12.5% purely dominant in one character and hybrid in the other, and so on. Recalling that hybrid zygotes produce somas with the dominant character, it follows that the progeny appear as follows :

<i>A. dom.</i> + <i>B. rec.</i>	18.75%
<i>A. rec.</i> + <i>B. dom.</i>	18.75%
<i>A. dom.</i> + <i>B. dom.</i>	56.25%
<i>A. rec.</i> + <i>B. rec.</i>	6.25%

A result which again agrees with experiment. The resulting mixtures of characters in tri to polyhybrids may be likewise predicted, by extending the principles already laid down.

BIBLIOGRAPHY.

- 1900** CORRENS, C. G. Mendel's Regel über das Verhalten der Nachkommenschaft der Rassenbastarde. *Berichte der deutschen Botanischen Gesellschaft*. XVIII. Jahrgang. Heft 4, pp. 158-168. May 23, 1900.
- 1900** DE VRIES, H. Sur la loi de disjonction des hybrides. *Comptes Rendus de l'Acad. des Sciences*. Paris. March 26, 1900.
- 1900b** DE VRIES, H. Das Spaltungsgesetz der Bastarde. *Berichte der deutschen Botanischen Gesellschaft*. XVIII. Jahrgang. Heft 3, pp. 83-90. April 25, 1900.
- 1900c** DE VRIES, H. Sur les unités des caractères spécifiques et leur application à l'étude des hybrides. *Revue générale de Botanique*. XII. pp. 257-271. July 15, 1900.
- 1889** GALTON, F. Natural Inheritance. New York. Macmillan & Co. pp. 259.
- 1865** MENDEL, G. Versuche über Pflanzenhybriden. *Verh. des Naturforscher-Vereins in Brünn*. Bd. iv, p. 1.

REGENERATION OF PROPORTIONATE STRUCTURES IN STENTOR.

T. H. MORGAN.

THE important results of Gruber and of Balbiani on the power of regeneration of pieces of *Stentor coeruleus* opened the way for further experiments; and the works of Johnson and of Lillie on the same form have added some further results of interest. There remained, however, one problem that had not been touched upon by these investigators, an answer to which is needed to make more complete our knowledge of the regeneration of unicellular forms. I refer to the question of the proportionate development of the new organs in pieces of different sizes, and from different parts of the body; and also the no less important question of the change in size of old organs that may be present on the piece at the time of its removal. It is the purpose of the present communication to describe certain experiments that bear on these questions.

Although it is evident, in a general way, from the figures given by Gruber and by Balbiani that a small piece produces a smaller peristome than does a large piece, yet their figures do not show definitely that such is the case, and, in fact, it would be difficult to determine that such is the case from observations made on the swimming animals. The figures that have so far been published represent the new stentor as it appears while contracted or when swimming. To obtain sufficiently accurate data for the problems that I wished to examine, it was necessary to make the measurements and drawings from the stentor at rest when in a fully expanded condition. The object of my work was to find an answer to the following questions: 1. Do small pieces produce a new organism having the typical proportions of the normal; and does it make any difference in this respect as to the part of the stentor from which the piece is taken? 2. If a piece containing the old

peristome is cut off, will it retain the old peristome, or absorb it and produce a new one of proportionate size? If the old peristome persists, will it decrease in size until it has assumed the typical proportions? 3. If a part only of the old peristome is left on a piece, will the missing parts be regenerated from it, or will a new peristome develop?

There appeared during January and February in one of the aquaria in the laboratory a large number of stentors, whose presence seemed to be connected with the appearance of vast numbers of vorticellas, on which they fed. The operation of cutting the stentor in two or more parts was carried out either by means of small scissors, or, in most cases, by a sharp scalpel. The latter operation is greatly facilitated by placing the animals in a dish of water, the bottom of which is covered by a layer of paraffin.

The following measurements give the length of the normal blue stentor and the greatest width of the peristome.

Length.	Width of Peristome.
2.8 mm.	.52 mm.
1.6	.46
1.4	.40
1.7	.50
1.7	.48
1.6	.44
1.9	.48

If a stentor is cut in two by a cross-cut, as indicated in Fig. 1, *A*, *a-a* (the anterior piece, *B*, being smaller than the posterior, *C*), the cut surfaces of each piece are closed almost instantly by the outer layer bending over the exposed part. Only a faint, clear line on the surface indicates where the cut has been made. The history of the anterior piece is as follows: In the course of an hour or two the piece becomes somewhat more pointed at the posterior end, and then fixes itself by a foot that appears at that end (Fig. 1, *B*). The posterior end now begins to draw out into a stalk, and after thirteen hours the piece has assumed the form shown in Fig. 1, *B*¹. The piece is still proportionately too broad for its length, for although the peristome has become reduced in size it is not as

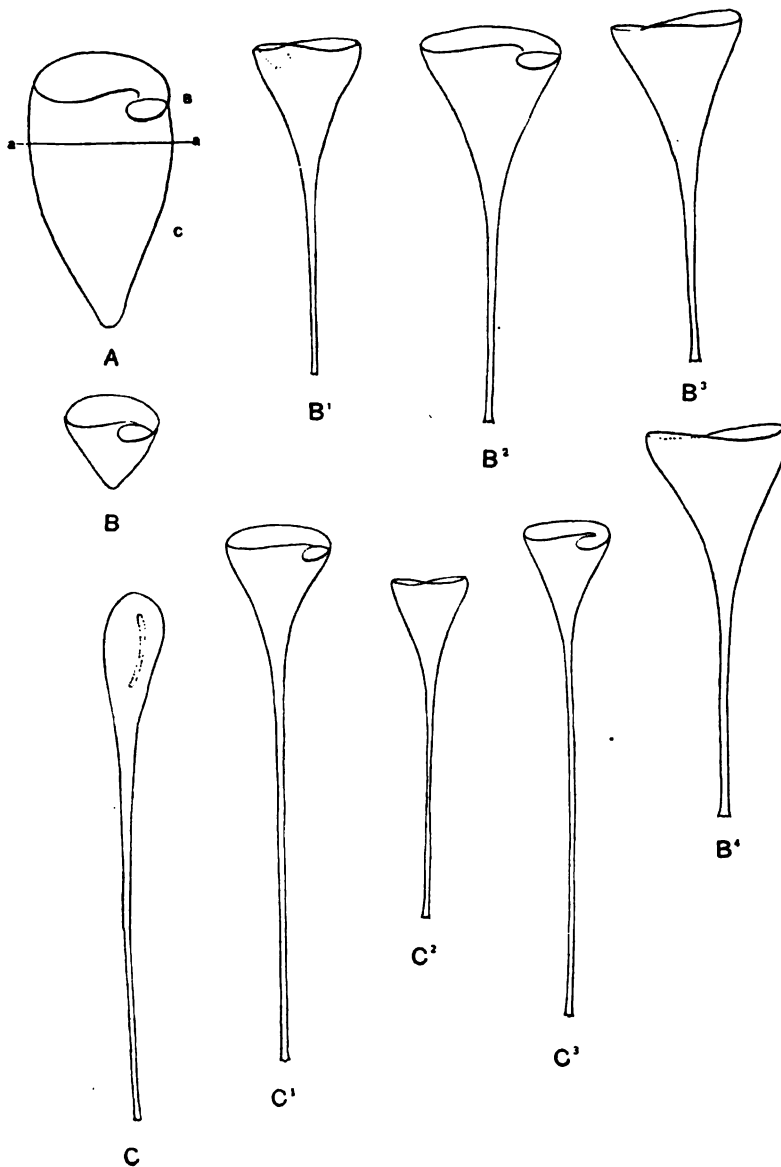


FIG. 1.—*A*, contracted stentor to show where the cut was made, *a-a*; *B*, anterior end five hours after the operation; *B*¹, an anterior piece thirteen hours after operation; *B*², *B*³, anterior pieces twenty-four hours after operation; *B*⁴, anterior piece forty-eight hours after the operation. *C*, posterior or foot end of *A*; *C*¹, posterior end five hours after operation; *C*², *C*³, posterior ends twenty-four hours after operation.

yet reduced sufficiently to give the piece the typical proportions. Two other head-pieces of this same series are shown in Fig. 1, B^2 , B^3 , that were drawn twenty-four hours after the operation. It is even more evident in these (compare with Fig. 2, A , for normal) than in the last that the peristome is too broad for the length of the stentor. Even after another twenty-four hours one of the pieces had still retained the same form as shown in Fig. 1, B^4 .

The posterior piece, C , fixes itself at once by the old foot, and may soon elongate to its full length. In the course of two or three hours a clear band appears extending somewhat obliquely over the rounded end of the piece (Fig. 1, C). Cilia appear along the band. In a few more hours, the rate depending on the temperature, the ciliated band moves forward around the anterior end of the piece, and in doing so bends around on itself into the characteristic peristome. A new peristome-field, or disk marked by delicate parallel lines, appears on the inner side of the band even before it moves forward, and as the band bends around to make the terminal peristome, the new disk comes to lie in its central part. A depression, that appears at the basal end of the band, forms the pharyngeal funnel. The new peristome is smaller than that of the original animal, and, as the figures show (Fig. 1, C^1 , C^2 , C^3), it is, in some cases, even smaller than the reduced peristome on the anterior piece. The foot-piece is also at first very long as compared with the size of the new peristome; and this condition may remain for several days.

These results demonstrate that, for some time after the new organs have developed, the new stentors retain some of the peculiarities of the part of body from which they have come. When the pieces are contracted, or are swimming, these relations are scarcely evident and might easily escape detection.

The transformation of an anterior piece into a new stentor is much more strikingly seen when only a small part of the anterior end is cut off. One set of observations on the same individual is represented in Fig. 2. The stentor fully extended is represented in Fig. 2, A . The anterior end had been cut off, as shown in Fig. 2, A^1 , while the animal was contracted.

After its removal the anterior end, *B*, contracted still more, so that its posterior cut-surface was quickly covered over by the bending in of the sides; the disk bulged forward. After a few hours the piece became somewhat pointed at its posterior end, and then fixed itself by a foot that appeared at the end.

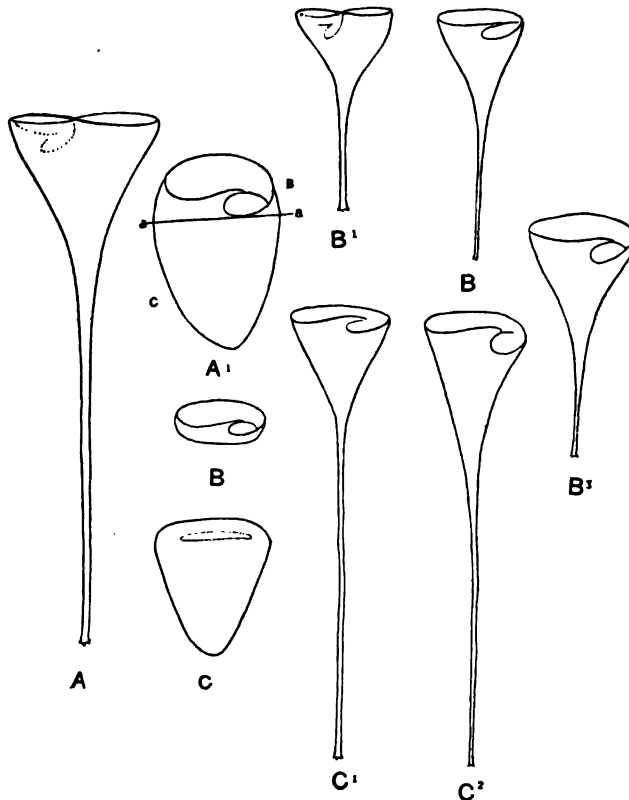


FIG. 2.—*A*, 1.4 × .4 mm. Stentor fully extended; *A'*, same contracted, cut in two at *a-a*; *B*, *C*, immediately after operation; *B'*, .56 × .26, anterior end after twenty-four hours; *B''*, .7 × .25, same after another forty-eight hours; *B'''*, .64 × .28, same after another four days, *i.e.*, seven days after operation; *C*, 1.2 × .27, posterior end after twenty-four hours; *C'*, 1.2 × .20, same after another forty-eight hours.

After twenty-four hours (it had been kept in the cold over-night, *i.e.*, for ten hours) it appeared as shown in Fig. 2, *B'*. The old stentor measured 1.4 mm. by .4 mm. This new stentor measures .56 mm. by .26. The old peristome has, therefore, decreased nearly to half its original width. Two

days later (Fig. 2, B^2) the stalk was somewhat longer, and after another four days (Fig. 2, B^3) the form had not materially changed. The development of the posterior piece of this same individual is shown in Fig. 2, C^1 and C^2 . The piece is about twice as long as the anterior piece, but its peristome is about the same size.

A similar operation was carried out on another individual; the results are shown in Fig. 3, A . A very small part of the

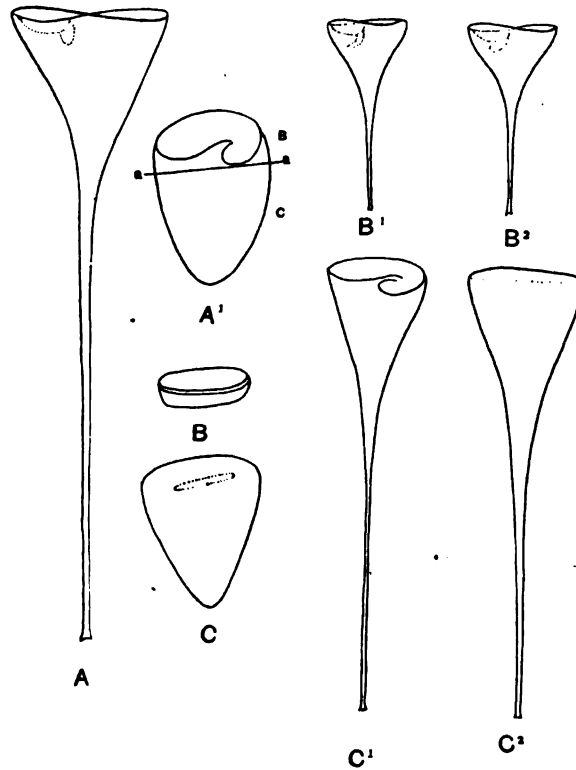


FIG. 3. — A , 1.7×4 . Stentor fully extended; A^1 , same contracted, cut in two at $a-a$; B , C , immediately after operation; B^1 , $.5 \times .21$, five hours after operation; B^2 , $.5 \times .24$, three days after operation; C^1 , posterior piece, $1.2 \times .27$, five hours after operation; C^2 , same, $1.6 \times .32$, three days after operation. In Figs. 2 and 3 one individual used in each. In Fig. 1 several individuals were used.

anterior end was cut off (Fig. 3, A^1). It contained, however, the entire peristome and disk (Fig. 3, B). About thirty hours after the operation the anterior piece appeared as shown in

Fig. 3, *B*¹. After three days more the new stentor had about the same form. The peristome is, as compared to that of the original stentor, too wide for the length of the new individual, although it is not much more than half the width of the old peristome.

The development of the posterior piece is shown in Fig. 3, *C*¹ and *C*². In this piece, particularly in the earlier stage, *C*¹, the peristome is smaller than was the original peristome, and also relatively smaller as compared with the entire length of the animal.

In order to be certain that the anterior pieces did not produce new peristomes during the night, they were kept in a cold place when not under observation; for I had found that under these circumstances the formation of a new peristome is greatly delayed, even although it may have begun to develop before the piece is subjected to the cold. In this way I could retard the development of the peristome for twelve hours, so that I felt certain that a new peristome had not developed on these anterior pieces in my absence.

In other experiments pieces of different sizes were cut from the foot-end in order to see if the size of the new peristome that is formed is in proportion to the size of the piece. It was found that a smaller peristome develops on a smaller piece, and a larger one on a larger piece; and this same relation holds also for pieces of different sizes for other parts of the body. It has been shown that cross-pieces from the anterior or posterior ends retain some of their original peculiarities even after the formation of a new individual, and that for several days the stentors from anterior pieces are too broad for their length, and individuals from the posterior end are too long for their breadth. Some of these newly regenerated stentors from the anterior pieces were kept for a longer period and supplied with food. Their measurements for from one to seventeen days after the operation are given in the following table. The measurements of three normal individuals of this lot were $1.6 \times .5$; $1.4 \times .4$; $1.1 \times .4$. The experiment began February 3.

ANTERIOR PIECES OF HALF SIZE OR LESS.

Feb. 4.	$\left\{ \begin{array}{l} .6 \times .22 \\ .85 \times .28 \\ .6 \times .23 \end{array} \right.$	Feb. 10.	$\left\{ \begin{array}{l} .85 \times .32 \\ .76 \times .28 \end{array} \right.$		
				Feb. 20.	$\left\{ \begin{array}{l} .96 \times .36 \\ 1.00 \times .39 \\ 1.04 \times .32 \\ 1.00 \times .40 \\ 1.00 \times .40 \\ 1.00 \times .39 \\ .9 \times .36 \end{array} \right.$
Feb. 6.	$\left\{ \begin{array}{l} .6 \times .23 \\ .6 \times .3 \\ .68 \times .2 \\ .7 \times .25 \end{array} \right.$	Feb. 13.	$\left\{ \begin{array}{l} .7 \times .34 \\ .8 \times .35 \times .25 \\ .9 \times .28 \times .2 \\ .64 \times .28 \\ 1.12 \times .39 \\ 1.2 \times .28 \end{array} \right.$		

It will be seen from this table that, after feeding, the stentors from anterior pieces grow larger. The increase takes place both in the peristome and in the length of the piece, so that the proportionate size of the disk to the rest of the piece remains about the same. The new stentors had begun to divide on February 13, and by February 20 there were about twice as many present as at first. They have, however, about the same proportionate size as at first. The question arises whether in the normal stentor the ratio of the breadth of the disk to the length of the pieces may not be less than in very large individuals. I measured some of the smaller individuals found in the aquaria with the larger ones and obtained the following results: $1.04 \times .36$; $.72 \times .26$; $.7 \times .28$; $.72 \times .3$; $.9 \times .38 \times .2$; $1.1 \times .28$; $.9 \times .36 \times .2$; $.98 \times .28$. There is seen to be some variation in the relative size of the length to the breadth; that is due in part to the individuals not always expanding to the same extent, and also in part to some of the measurements of the peristome not having been made in the widest part, but there are also actual differences, as some very careful measurements have shown. It will be seen that while in large stentors the greatest breadth of the peristome is about one-fourth, or nearly so, of the total length, in the small individuals the breadth is more nearly one-third of the length; therefore the peristome is proportionally somewhat larger for smaller pieces. Comparing these measurements with those of the sizes of individuals derived from pieces of the anterior end, we see that they have reached in several cases the characteristic form *for a small individual*. Since there is

a good deal of variation in the proportion between the width of the peristome and the length of the animal both for small normal individuals and for those that have come from anterior pieces, it may be stated that pieces from the anterior end may produce new stentors whose proportions come within the range of variation of size of normal small stentors of about the same length. The measurements of posterior pieces, that are at first too long for the size of the peristome, show that they, too, assume more typical proportions. Thus one of the posterior ends of the last series measured, on February 6, $1.0 \times .25$. On February 10, two other individuals in the same list measured $1.4 \times .38$; $1.2 \times .4$. The peristomial region had, therefore, reached the full size.

A somewhat crude comparison may bring the results home. If a man were cut in two at the waist and the pieces behaved in the same way as those of stentor, two new individuals would develop. The anterior half would produce a small man with a head too large for his height, *i.e.*, his legs would be too short for a man with that sized head. Although the old head had grown smaller, it would be still too large for the rest of the new man. In fact, his proportions would be more like those of a baby whose head is relatively too large for his length as compared with that of a man, and his legs too short. It is just this result that we have found for anterior pieces of stentor. If the new man were supplied with food, all parts of the body would grow larger; but as he got larger his legs would grow faster than his head.

The posterior end of our imaginary man would have at first legs too long for his total length, and his new head would be relatively too small; but if he were fed his head, shoulders, and arms would grow faster than any other part and continue to grow until the proportionate size had been reached. If he were not fed, it is possible that his head and upper part might increase more slowly in size at the expense of the material in his legs, and the latter would get smaller until a balance was reached. The result would be that a boy rather than a baby was produced.

In other experiments pieces were removed that contained

only a part of the peristome. In one series these pieces were cut off, as shown by the line *a-a* in Fig. 4, *A*; so that there was a smaller and a larger piece, the former, *B*, containing a part of the old peristome, but not any part of the pharyngeal funnel, and the latter, the larger piece, *C*, containing also a

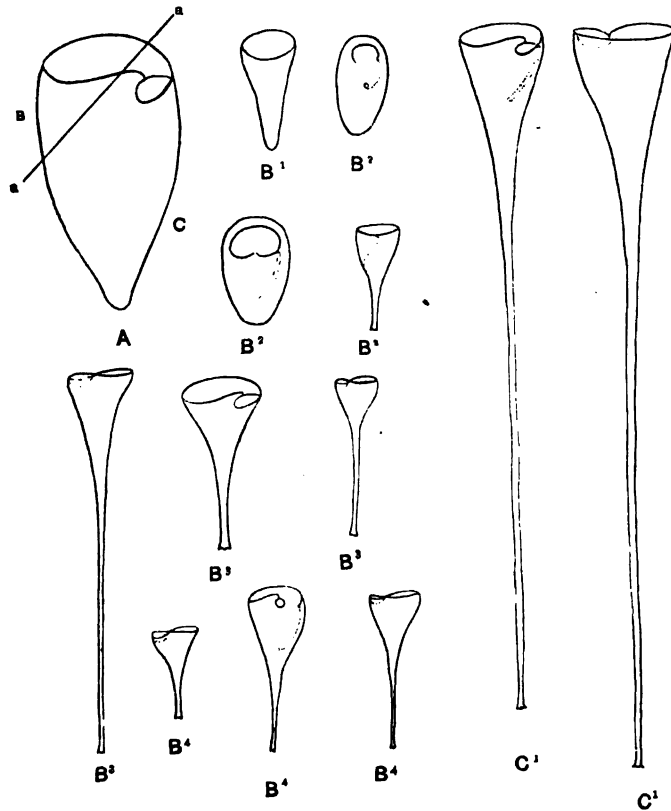


FIG. 4. — *A*, stentor partially contracted, cut in two at *a-a* into small piece, *B*, and larger, *C*: *B*¹, piece *B* after seven hours; *B*², *B*², *B*², piece like *B* after twenty-four hours; *B*³, *B*³, *B*³, same after twenty-nine hours; *B*⁴, *B*⁴, *B*⁴, same in part after forty-eight hours (after operation). *C*¹, *C*¹, piece *C* after seven hours.

part of the peristome as well as the funnel. The cut surface of each piece is quickly closed by the bending in of the sides, and the cut ends of the peristome are generally brought together to make a closed ring. A foot develops on the basal end of the anterior piece, *B*, *B*¹, and a stalk is soon produced in that region. This piece may remain for twenty-four hours in this

condition. Sooner or later a new ciliated band appears on the old wall behind the part of the old peristome, as shown in Fig. 4, B^2 . The band moves forward, fusing with the old ring at one point and, replacing the latter, produces a new peristome. Whether the piece of the old ring is entirely obliterated, or whether a part of it remains to contribute to the new peristome, I did not determine. The regeneration of a new peristome on these pieces may be delayed for several days (Fig. 4, B^4), and, in general, does not appear as soon as on pieces that do not contain any part of the old peristome. Five or six series of experiments of this sort, each series of a number of pieces, were made, and the smaller pieces followed with great care. Only those smaller pieces were isolated that contained no part of the old funnel. Nearly all the pieces behaved in the way just described, but in one or two a small funnel developed where the cut edges came together. This may have been due to a very small piece of the original funnel having been cut off, or to the piece having come from very near to the old funnel, or, as seems more probable, to the development of a new funnel from the old ciliated band. If the last interpretation is correct, it shows that in exceptional cases the peristome may complete itself. In the large majority of cases this does not occur and a new peristome and funnel develop at the side and move forward. In nearly all cases the cut ends of the old peristome come together, meeting in a slight notch. In one or two instances one band lay slightly below the other at the meeting point, producing a peristome exactly like the normal in shape, only the funnel was absent. If a small piece of the old funnel is left, it assumes the characteristic position of the funnel, and, in fact, becomes such to all appearance, although this peristome is generally replaced later by a new one.

Gruber studied the regeneration of pieces somewhat similar to these without a funnel, and states that the remaining part of the old peristome gives rise to a new one, but I have not found this to be the case. If the piece is kept under close observation, the development of a new peristome is found to take place in the way just described. The change is sometimes so rapid that a few hours may suffice to bring it about.

The history of the complementary piece (Fig. 4, *C*) is as follows: After the cut surface has been closed over and the edges of the peristome brought together, the piece may immediately fix itself by the old foot. The piece elongates to its full length, which is the same as that of the former animal (Fig. 4, *C*¹). In some of these pieces I have observed the development of a new peristome in the course of a few hours after the operation (Fig. 4, *C*¹). It seems that this takes place sooner when only a small part of the old peristome and funnel is left than when a larger part remains. In cases in which a large part of the old peristome remains a new peristome may not develop for several days; and in some cases I have not found it to appear at all, but I cannot state positively that it does not ultimately appear. Since even normal individuals may produce a new peristome, the appearance of a peristome on these new stentors after several days may be only the regular process of renewal of that organ. In two cases, in which a new peristome appeared after two days, the old one had begun to break down while the new band was developing. In all other cases the old part was still active and normal in appearance up to the time of its replacement by the new ciliated band. These results show that even in a large piece the new peristome is not regenerated from the old one. The presence of the old pharyngeal funnel in these pieces does not make any important difference in the end result, although it may be that pieces of this sort regenerate less quickly than when the piece does not contain the funnel portion of the old peristome.

Another experiment that supplements the preceding one in several respects consists in cutting the stentor in two, as indicated by the line *a-a* in Fig. 5, *A*. In this case the smaller piece, *B*, contains the funnel part of the peristome, while the larger piece, *C*, contains the remaining part of the peristome.

The smaller piece, *B*, closes in, develops a foot, becomes attached and produces a stalk. The edges of the peristomial ring unite more or less, as shown in Fig. 5, *B*¹. In one case a new peristomial band appeared six hours after the operation, moved forward, and produced a new peristome (Fig. 5, *B*¹). In

other cases, in which the piece was small in comparison to the size of the remaining part of the peristome, a new peristome did not appear in one case until after thirty hours (Fig. 5, B^4); in other cases a new peristome had not appeared at this time (Fig. 5, B^8).

The complementary piece (C , Fig. 5) closed in, fixed itself, and extended to its full length. In pieces in which the

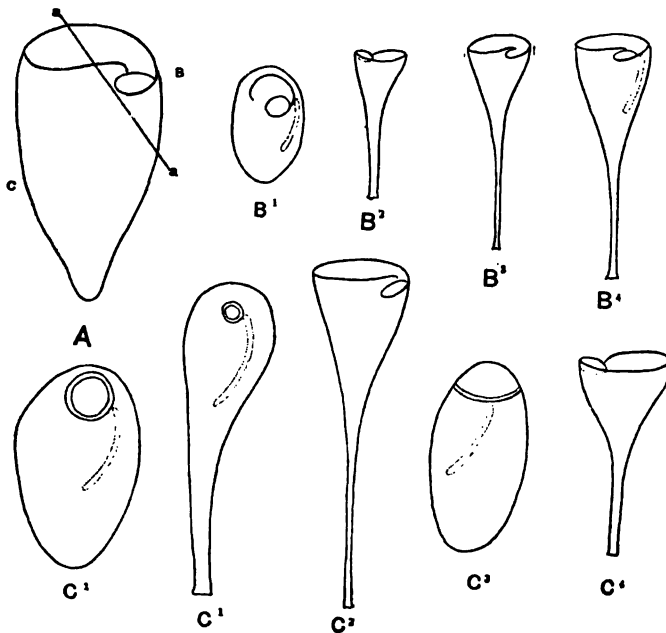


FIG. 5. — A , stentor partially contracted, cut in two at $a-a$ into a small piece, B , and a large piece, C ; B^1 , piece like B after seven hours; B^2 , piece like B after twenty-seven hours; B^3 , B^4 , piece like B after twenty-nine hours; C^1 , C^2 , piece like C after seven hours; C^3 , C^4 , piece like C after fifty-one hours.

remaining part of the peristome, that had united to make a ring, was quite small a new ciliated band appeared in four hours; in others, in six hours (Fig. 5, C^1 C^1); and in pieces with a larger peristomial region, after twenty-four and even after fifty-one hours (Fig. 5, C^3). It is interesting to note that in these pieces the region from which in the normal individual the peristomial band is formed has been more or less completely removed, yet a new peristomial band may very quickly

appear. I did not attempt to determine the position of the new band in its relation to the region of closure of the piece.

In several cases in the last two experiments, and in some other experiments like those shown in Fig. 6, *A*, *B*¹, *C*¹, small pieces were sometimes cut off that contained a part of the old peristome, but which did not fix themselves, or assume the characteristic form. As these pieces were generally small, although not below the minimal size, there can be little doubt that most of them did not contain any part of the nucleus, and in several cases I proved this to be the case by staining the pieces in picro-carmin. The result shows that in the absence of the nucleus a piece containing a part of the old peristome cannot complete the peristome from the remaining part. This is the

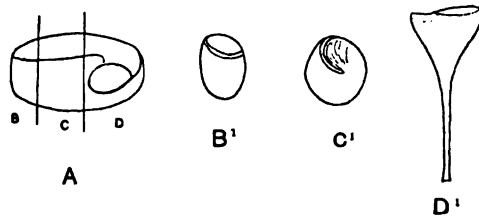


FIG. 6. — *A*, anterior end of stentor cut off and then divided into three pieces. Two of these, *B*¹, *C*¹, were apparently without nuclei, and did not produce a new peristome or assume the typical form.

less to be expected since it has been shown that even in nucleated pieces the new peristome is produced not by the old one, but by the development of a new peristomial band. The result is interesting in connection with a result obtained by Gruber, *viz.*, that if a non-nucleated piece containing a part of the newly forming ciliated band is obtained it produces from the band a new peristome. My results show that a piece of the old band cannot act in this way. That the presence of the nucleus is connected with the formation of a new peristomial band seems highly probable, but I can easily imagine that could a non-nucleated piece be supplied with certain unformed elements it might be capable of producing a new peristome. The results do not seem to me to show more than that the nucleus supplies certain products of metabolism that must be present before the protoplasm can successfully carry out its

innate tendency to complete the typical form. We are not justified, I believe, in drawing the conclusion, as Gruber has done, that preformed elements of the peristome exist in the nucleus and must be set free in order to initiate the development of a new peristome.

Lillie has found that the smallest piece of *Stentor polymorphus* that becomes a perfect form is equal to a sphere of about $80\ \mu$ in diameter. The average size of the stentors was equal to $230\ \mu$. This makes the volume of the smallest stentor about $\frac{1}{27}$ of the normal. For *Stentor coeruleus* the smallest stentors measured $90\ \mu$ ($=\frac{1}{11}$ mm.), the average normal stentor $280\ \mu$ ($=\frac{7}{5}$ mm.). Therefore the former is about $\frac{1}{27}$ of the latter.

Although I have not worked specially on this problem, yet I have obtained some small stentors that were proportionately smaller than those obtained by Lillie. Thus one individual measured when extended $.25 \times .08$ mm., and when contracted into an oval or nearly into a sphere $.08 \times .08$ ($=\frac{1}{12}$ mm.). The larger normal stentors measured about $.4 \times .32 \times .32$ when contracted. Although it is only possible to give a general estimate of the relative size of these two individuals, the smaller cannot be over $\frac{1}{81}$ of the former. It would be a mistake to infer from this, as well as from Lillie's calculations, that the latter came from a piece $\frac{1}{11}$ or even $\frac{1}{27}$ of the original stentor. The protoplasm of stentor is so vacuolated that a piece losing the fluid in the protoplasm might become much smaller than when first removed.

Lillie states that he believes that it would be possible to obtain a smaller individual of *S. coeruleus* than $\frac{1}{11}$ mm. The one that I obtained was in fact somewhat smaller, *viz.*, $\frac{1}{12}$ mm. The difference in our results depends, therefore, rather on the size of the normal average stentor with which the comparison is made than on the smallest individual obtained. Lillie says that he does not think there can be much difference in the absolute size of the smallest stentors, whether one uses the largest or the smallest normal specimens. It seems to me that this may or may not be true, according to what factors may enter into the result.

The conclusion that a piece $\frac{1}{27}$ or even $\frac{1}{64}$ of the entire animal can produce a new individual can give only a most general idea of the relative size of the smallest piece, since more depends on the size of the normal individual than on that of the smallest pieces, and there is for stentor a very wide range of size that may be called normal. A large normal individual may contain eight times the volume of a small normal individual. More significant, therefore, is the absolute size of the smallest piece capable of regeneration, and in this respect my results are practically in accord with those of Lillie.

Several experiments were made in which pieces were cut in two longitudinally. In a longer or shorter time most of the halves produced a new peristomial band that became a new peristome. As this experiment did not give promise of much that is new, it was tried in only a few cases.

A few casual observations made during the course of the work may be briefly mentioned. The stentors were observed dividing on several occasions, but Johnson's excellent figures and account of these stages leave nothing new for me to add. I have often noticed that after division the two products are found attached side by side, and if they are not disturbed a little colony may arise in the same spot. Several times I have observed that two individuals that have been formed by division of one of the regenerated posterior pieces were unequal in size, although I do not know whether the smaller individual was the distal or the proximal one.

As Gruber has pointed out, the first steps in the process of division and of regeneration are the same, and this holds also for the physiological replacement of the old peristome. In all cases a peristomial ciliated band appears *on the side* and moves forward around the anterior end to become the new peristome. We have here another illustration that shows that during the process of regeneration the factors that appear in the normal growth may take part in the regeneration, and this relation appears to hold for unicellular as well as for multicellular forms.

In many cases, especially where a somewhat oblique cut has been made, the superficial blue stripes come together over the

cut surface in a most irregular way, yet this does not appear to interfere with the subsequent regeneration ; and after a time the stripes appear to be more regularly arranged. That a certain amount of absorption takes place, and possibly also development of new stripes, seems probable, but I have not studied these changes in any detail. It would be interesting to find out if in cross-pieces of the body the number of the stripes remains the same and their size becomes smaller, or whether the number of the stripes is proportionately reduced.

On several occasions I have tried to graft together pieces of different stentors, but the exposed surfaces close so quickly that I have not been able to get the pieces to unite. It does not seem altogether improbable that the result could be brought about by cutting two stentors at the same time, one about the other. A lucky cut might bring two exposed inner surfaces together, and they might stick to each other, but so far I have not been able to carry out successfully this experiment.

In a few cases the stentor was split partially in two pieces, but generally the halves soon fuse together. It is of some interest to find that, although the peristome was cut in two and had reunited, a new peristome was not produced, showing that the operation alone does not initiate the changes that lead to the development of a new peristome.

The development of a new peristome in a piece that contains a part of the old one appears to be due to the lack of proportion between the old part (even when it contains all the essential parts of the peristome) and the rest of the piece. This result is unique, since in all other forms in which a part of an old organ remains the new organ regenerates from that part. In stentor this does not occur, but a new organ is produced. It is important to observe, however, that this is the characteristic way in which stentor produces a peristome, so that the organisms make use of a process that already exists. The reduction in size of the old peristome in pieces from the anterior end is the result that has most interested me. It seems to be due to the withdrawal of material from the anterior region to form the body and stalk of the new stentor.

The change in shape of the piece, *i.e.*, the production of the typical form, is primarily the result of a shifting of the material that carries with it a loss of material in the old part. Other so-called formative factors may have some share in the reduction in size of the old peristome to one proportionate to the rest of the piece, but the simple loss of material will, I think, account for the greater part of the change.

What primarily brings about this change in the material so that the typical form is produced is a question to which at present there is no answer.

BRYN MAWR, March 12, 1901.

ABSTRACTS OF PAPERS

PRESENTED AT THE MEETINGS OF THE

AMERICAN MORPHOLOGICAL SOCIETY

AT BALTIMORE, DECEMBER 27 AND 28, 1900.

I. FISSION AND REGULATION IN STENOSTOMUM LEUCOPS.

C. M. CHILD.

THE single individual of *Stenostomum* differs considerably in size, according to conditions. Well-nourished specimens may reach a length of nearly one and one-half millimeters, while specimens measuring only one-half millimeter are often found when food is scarce. The length is about eight or ten times the transverse diameter.

The animals usually occur in chains, the number of zoöids varying from two to nine. Ordinarily chains do not consist of more than five zoöids, the uneven number being due to the fact that the anterior zoöid precedes the others in division. The short chains of two or three zoöids occur when food is scarce. In well-nourished specimens the fissions succeed each other more rapidly, and longer chains are the result. Each particular septum occurs, with little variation, in a definite, characteristic position, this apparently being determined largely by the relative degree of development of the two ends of the zoöid which it divides.

Entodermal tissue is necessary for regeneration. Portions containing all the other tissues of the body except entoderm

do not regenerate, but if a small portion of the entodermal digestive sac be present regeneration is complete, provided the piece is above a certain size.

If a chain be artificially separated into its zoöids before they have attained their full development, each zoöid undergoes a form-regulation, assuming within a few hours what may be called the normal proportions, *i.e.*, the length becomes eight or ten times the transverse diameter. This regulation does not occur while the individual is a zoöid in a chain, because the whole chain is not simply a series of individuals, but in some degree a single individual, and therefore possesses certain proportions differing from those which each zoöid would possess if single.

When the chain is cut at various points between the zones of fission, the results differ according to the degree of development of the particular zone of fission concerned and the parts adjoining it. If a piece containing a very young septum be cut out from a chain, the septum disappears and a single perfect individual is formed from the parts, which originally belonged to two different zoöids. A head is regenerated at the anterior cut surface, a tail at the posterior end. If the included septum be more fully developed, it remains, and the part anterior to it is completely absorbed by the part posterior to it, the head of the new individual resulting being the head which was forming just posterior to the septum. The relative size of the parts anterior and posterior to the septum does not affect the result, unless the posterior piece be very small. It is always the anterior part which is absorbed, never the posterior. If the septum be still more advanced in development, the portion anterior to it is only partly absorbed. It regenerates a new head and becomes a perfect zoöid, though at first it decreases in size, owing to the partial absorption.

In general each zoöid tends to absorb material from the zoöid anterior to it. Each zoöid, however, offers a certain resistance to this absorption, the resistance increasing as it approaches the condition of independent individuality. When the individuality of a zoöid is destroyed or reduced to a lower

degree, *e.g.*, by cutting it in half, the posterior half may be completely absorbed by the zoöids posterior to it.

The sexual individual arises as a zoöid in a chain, but when the sexual organs appear, asexual reproduction ceases. The single sexual individual may, however, attain a length equal to that of the longest chains. The power of regeneration is much less in the sexual than in the asexual condition. Apparently in the former the energy is chiefly directed toward the elaboration of the sexual products.

II. THE OCCURRENCE OF *GUNDA SEGMENTATA* IN AMERICA.

WINTERTON C. CURTIS.

A SPECIES of *Gunda*, which in its external features seemed identical with the *G. segmentata* of Lang, was found in large numbers at Sandwich on Cape Cod. The internal arrangement is not, however, as regular as Lang describes for *G. segmentata*. From a comparison with Verrill's figure of *Procerodes ulvae* collected in the same region (*Trans. Conn. Acad.*, Vol. VIII, January, 1893), it is probable that the two forms are identical and that Verrill has figured the head incorrectly.

III. SOME DISPUTED POINTS IN THE ANATOMY OF THE LIMPETS.

M. A. WILLCOX.

THE following points were made:

1. The space previously described by Willcox as the nephridium is lined throughout by more or less columnar cells provided with long, delicate cilia and loaded in the fresh condition with dark green granules. The histology lends no countenance to Haller's contention that the posterior part of this sac represents coelom, the anterior nephridium.

2. The space which in most species of *Acmaea* underlies the viscera on the left side in *A. testudinalis* stretches almost across the body, and lacks entirely the ciliated cells characteristic of the nephridium. This negatives the opinion that the space in question is a paired structure whose fellow of the right side is represented by the posterior part of the nephridium.

3. A subradular organ, whose presence in the *Docoglossa* has been denied, exists in both *A. testudinalis* and *A. fragilis*. It is situated on the underside of the odontophore, just behind the tip of the radula, and is a triangular, somewhat cushion-shaped organ, divided by a V-shaped groove into an anterior and a posterior part. The posterior part is marked by transverse grooves and is covered by tall, columnar epithelial cells, some of which seem to be ciliated, while others are somewhat fusiform and have much the appearance of sense cells. The innervation has not been traced, but no ganglia are to be found in the organ. The subepidermal portion consists of connective tissue with scattered and inter-crossing muscular fibers.

IV. THE HABITS AND LIFE HISTORY OF ARGULUS WITH REFERENCE TO ITS ECONOMIC IMPORTANCE.

CHARLES B. WILSON.

IN the town of Warren, Mass., is a small pond which was stocked with carp and bass several years ago. The fish seemed to thrive well until the fall of 1899, when they began to die off in considerable numbers, with no apparent signs of disease or injury. No clue to the cause of the devastation could be obtained till the spring of 1900, when several suckers were speared in the outlet of the pond whose gill chambers were full of the parasitic copepod *Argulus*, probably *A. cato-stomi*. The gentleman who owned the pond stated that these copepods were common on most of the fish caught there, and his statement was afterward verified. On being put in an aquarium with dace, roach, and bream, they attacked these fish

viciously and the next morning they were found dead. *Argulus* deposits its eggs instead of carrying them around like other crustaceans, arranging them in rows on sticks, stones, etc., with their long diameters parallel.

When laid they are covered with a jelly envelope consisting of beads of jelly arranged in rows parallel with the long diameter of the egg. These harden into a brittle shell. The eggs are fertilized outside the body of the female and there is no copulation. The egg hatches into a typical nauplius, which after one or two moults changes into a metanauplius having highly developed clasping organs in the shape of barbed claws terminating the second maxillae. On putting two small dace into the aquarium with about two hundred of these larvae, the latter made no attempt to use their claspers, but the fish, on recovering from their fright, ate up every one of the larvae. On inquiry it was found that the pond in question had been seined for three years, and that the dace and roach had been sold for pickerel bait. *First conclusion*, the subject of parasitism is not so one-sided as would appear at first sight. *Second conclusion*, the protection of small fish like dace and roach in our fish ponds may be one of the best preventives against such parasites as these.

V. THE ANATOMY AND DEVELOPMENT OF THE POSTERIOR VENA CAVA IN DIDELPHYS VIRGINIANA (KERR, LINN.).

C. F. W. McCLURE.

AN examination, thus far, of forty-eight opossums has brought to light many interesting variations concerning the mode of formation of their posterior vena cava.

These variations are so pronounced and so closely accord with certain embryonic conditions described by Hochstetter for *Echidna aculeata*, it seems to the writer as not improbable that the development of the posterior vena cava may take place in *Didelphys* and *Echidna* in substantially the same manner.

In all marsupials hitherto examined (*Petaurus taguanoides* excepted) the posterior vena cava has been found to lie ventral to the aorta between the renal and common iliac veins, and to be formed through a union of the common iliac veins, which takes place *ventral* to the arteries.

In *Didelphys virginiana* the posterior vena cava is not formed in this manner.

In fact, the mode of formation of the posterior vena cava was found to be so variable in *Didelphys virginiana* that it is quite impossible to assign any one mode of origin for this vessel which may be regarded as typical of the species.

For descriptive purposes the various modes of origin of the posterior vena cava in *Didelphys* have been classified by the writer under three main types as follows :

Type I includes those cases in which the internal iliac veins unite with the external iliac veins to form the posterior vena cava, *ventral* to the common iliac arteries or *ventral* to the aorta.

Type II includes those cases in which the internal iliac veins unite with the external iliac veins to form the posterior vena cava, *dorsal* to the common iliac arteries or *dorsal* to the aorta.

Type III includes those cases in which the internal iliac veins unite with the external iliac veins to form the posterior vena cava, both *dorsal* and *ventral* to the common iliac arteries or both *dorsal* and *ventral* to the aorta.

So many variations of this type were met with that a further subdivision of Type III was found necessary, as follows :

Type III, A, includes those cases in which the principal union between the internal and external iliac veins lies *ventral* to the arteries in question.

Type III, B, includes those cases in which the principal union between the internal and external iliac veins lies *dorsal* to the arteries in question.

Type III, C, includes those cases in which the above-mentioned *dorsal* and *ventral* unions are *sub-equally* developed.

The following table shows how the above-mentioned types were distributed among the forty-eight individual opossums examined (twenty-four males and twenty-four females).

TYPE.	♂	♀	TOTAL.	
Type I	5	9	14	14
Type II	6	7	13	13
Type III				
<i>A</i>	3	2	5	21
<i>B</i>	8	5	13	
<i>C</i>	2	1	3	
TOTAL	24	24		48

A comparison with the development stages in *Echidna aculeata* shows, I think beyond the question of a doubt, that the variations in the method of formation of the posterior vena cava in *Didelphys*, so far as its posterior tributaries are concerned, are modifications of a ground plan arrangement similar to that described by Hochstetter for his *Echidna* embryo No. 45.

The writer's investigations upon the development of the posterior vena cava are as yet incomplete. So far as they have gone, however (an examination of five 15-millimeter embryos), they decidedly favor the above conclusions.

VI. THE CROSSING OF THE OPTIC NERVES IN TELEOSTS.

G. H. PARKER.

IN ten species of symmetrical teleosts, in each of which one hundred specimens were examined, the right optic nerve was dorsal at the crossing about as frequently as the left. The two types of crossing (right nerve dorsal and left nerve dorsal) were not correlated with sex and were about equally frequent

in specimens taken from one school of fish. In one hundred specimens of the winter flounder (*Pseudopleuronectes americanus*), whose eyes are on the right side, all had the left nerves dorsal. In seventeen specimens of the summer flounder (*Paralichthys dentatus*), whose eyes are on the left side, all had the right nerves dorsal. In one hundred specimens of the stellate flounder (*Platichthys stellatus*) all had the left nerves dorsal, notwithstanding the fact that fifty of these fish had their eyes on the right side and fifty on the left. Although each species of symmetrical teleosts examined showed about equal numbers of the two possible types of optic nerve crossing, the flounders showed only one type for each species.

VII. A NEW TYPE OF BUDDING IN ANNELIDS.

H. P. JOHNSON.

Two gigantic undescribed species of Pacific coast *Syllidae* produce reproductive zoöids by collateral budding from a definite proliferating region near the posterior extremity.

The single specimen of the larger species (*Trypanosyllis ingens*, sp. nov.) at my command was too poorly preserved for thorough study, but what I have learned about its budding agrees essentially with the fuller knowledge acquired from the other species. The buds numbered about thirty, all turgid with nearly ripe ova. No very young buds were detected, as in the succeeding species.

Of the other species (*Trypanosyllis gemmipara*, sp. nov.) I have several specimens, but only one with buds. They develop from a proliferating region twenty somites anterior to the pygidium. The advanced buds are broadly elliptical, and much flattened dorso-ventrally. Each is attached at its head-end by a short pedicle. The somites number 20-28, with parapodia which are miniatures of those of the parent. The prostomium has large eyes, a pair of antennae, and brain. There are a pair of ventral nerve cords, a muscular system, septa, and large paired masses of sperm cells in every coelomic chamber from

prostomium to pygidium. Purely vegetative organs (*e.g.*, mouth, alimentary canal, anus, and nephridia) are absent, although a rudiment of the alimentary canal may exist as a median strand of tissue extending the length of the bud.

The youngest buds form a cluster of about twenty-five attached to the right side of the zone of proliferation, on its ventral aspect. The earliest-formed organs are the anal cirri, at first two distal protuberances which elongate and become moniliform before the bud segments. Apparently the bud contains only ectoderm and mesoderm, which are continuous with the same germ layers of the proliferating region.

In neither species are there any reproductive cells in the body of the parent anterior to the proliferating region, but sperm cells are present in *T. gemmipara* in the twenty parental somites back of this point.

VIII. AMPHIBIAN STUDIES.

J. S. KINGSLEY.

THE following are the chief points made in the paper :

1. The *Salamandrina* form the central Urodele stem, and the Perennibranchs and Derotremes have been derived from this stem by degeneration and the retention of larval characters.
2. The Urodeles cannot have been the ancestors of the *Anura* ; the anuran tadpole resembles the Urodele only in superficial characters ; the *Anura* have descended directly from the *Stegocephala*.
3. *Amphiuma* has no tentacular apparatus at any stage ; what was described as such by Davison was a trematode parasitic in the suborbital blood vessel.
4. The Caecilians differ from all Urodeles in the fact that the palatine nerve receives a branch from the ophthalmicus profundus instead of from the maxillaris superior nerve.
5. The Caecilians have not descended from the Urodeles, nor is *Amphiuma* a neotaenic Caecilian. The Caecilians are degenerate in loss of limbs and tail ; in all other respects they are the most primitive of living *Amphibia*.

6. No *Stegocephalan* as yet known can have served as ancestor of *Urodeles*, *Anura*, or *Caecilians*. The parent form must have possessed characters intermediate between the known *Stegocephali* and the *Crossopterygian* ganoids.

7. The ancestry of the *Amphibia* must be sought in the *Crossopterygii* and not in the *Dipnoi*.

8. The balancer of the *Urodele* larva is a modified external gill belonging to the hyoid arch.

IX. PHAGOCYTOSIS IN A MAMMALIAN OVARY.

MAYNARD M. METCALF.

It has long been known that in the ovaries of certain Mammals and Fishes syncytia of young ova are found, and that of the several nuclei in such a syncytium one or but few persist as nuclei of definitive ova. Apparently, in these cases, the cells which disappear are used as food for the persistent ova.

In the ovary of the common Cat somewhat similar conditions have been observed by the author. Many of the young ova, in the stage when they are surrounded by a follicle consisting of but two layers of cells, are seen to have ingested many of the follicle cells, and the nuclei of these ingested cells can clearly be seen, some quite perfect (newly ingested), others apparently in different stages of digestion. The nuclei of these ingested cells, when almost completely digested, appear as groups of granules, these granules being apparently the remnants of the nodal thickenings of the chromatin network of the ingested cells. Such an ovum with its ingested nuclei very closely resembles the young blastomeres of a *Salpa* embryo, which have the same habit of devouring follicle cells.

Many young ova with ingested follicle cells were found in one Rat ovary. In another ovary of a White Rat no ingestion of follicle cells was found, nor was ingestion found in the ovary of a Gray Squirrel examined. Pressure of other duties has prevented the author from determining if such ingestion of follicle cells be normal in the ova of Rats and, if so, what

relation, if any, it may have to the age of the individual or to its condition as regards reproduction. The observations are reported in the hope that other observers may have them in mind. If the phenomena are at all general among *Mammalia*, they should be seen in many laboratories in the usual histological demonstrations.

Similar phenomena are, of course, well known in several groups of *Invertebrata*.

X. THE MAMMALIAN LOWER JAW.

W. H. RUDDICK AND J. S. KINGSLEY.

IN no adult mammal, recent or fossil, is the lower jaw known to consist of more than a single bone, and no author, save W. K. Parker, whose observations appear to have been overlooked, has shown the existence of more elements in its development. We are able to confirm Parker in his account and to identify in the mammals the following bones of the non-mammalian groups: (1) articulare, (2) angulare, (3) splenial, (4) dentary. Of these, articulare and angulare unite to form the malleus, while the definitive lower jaw is composed of dentary and splenial. Two cartilages participate in the formation of the lower jaw, — the Meckelian and a second larger cartilage lying external to this, which, like Parker, we homologize with one of the lower labials of the *Ichthyopsida*. In its ossification this cartilage is strikingly similar to amphibian cartilages, and the resulting bone — a part of the dentary — gives rise to the posterior part of the lower jaw, including the coronoid and articular processes. In the existence of this lower labial is to be found the explanation of the shifting of the articulation of the lower jaw. It is noteworthy that a lower labial occurs in about the same position in the ganoid *Polypterus*.

XI. AN APPARATUS IN THE CENTRAL NERVOUS
SYSTEM OF VERTEBRATES FOR THE TRANS-
MISSION OF MOTOR REFLEXES ARISING
FROM OPTICAL STIMULI.

PORTER EDWARD SARGENT.

IN *Amia*, at about the time of hatching, there arises in the anterior portion of the roof of the optic ventricle a group of cells, eighty to one hundred in number, formed by the differentiation of indifferent neuroblasts. During the first and second days of larval life the axons develop from these cells as exceedingly fine processes, growing directly toward and into the optic ventricle. Early in the third day the adjacent axons come together in groups and coalesce at their tips, in their further growth through the cerebro-spinal fluid appearing as a single fibril. Later these fibrils coalesce with others similarly formed, and in their growth posteriorly through the ventricles and canalis centralis form what has been known as Reissner's fibre, which is then a fibre tract made up of many axons closely united and surrounded by a single medullary sheath. Through the posterior portion of its course there come off from it fine fibrils which pass through the canal obliquely backwards and enter the tissue of the cord.

In the first day after hatching there may be found in the extreme posterior end of the canalis centralis a number of small cells, three to four micra in diameter, lying in the lumen of the canal and ventriculus terminalis. Some eight to ten of these cells persist and continue to develop. Increasing rapidly in size, they become spindle-shaped and send their axons cephalad through the canal. The axons are at first separate, but later coalesce as they grow forward, and, eventually meeting the system of axons from the cells of the tectum growing posteriorly, the two interweave in a way not yet clearly made out.

The development of this apparatus in *Amia* is typical of its development in all vertebrates, though in some groups

there are considerable variations. In the Skate the cells are of conspicuous size, and three to four hundred in number. They are multipolar, sending processes to the ectal portion of the tectum, where they come in direct contact with the endings of the optic nerve fibres and give rise to two fibre tracts, one of which passes posteriorly to the cerebellum, the other anteriorly and out into the optic ventricle to form the Reissner's fibre. In reptiles and birds the apparatus appears at a late stage and is not fully established until just before hatching.

In *Petromyzon* this apparatus is not fully established until the second month of larval life. The cells, about twelve in number, form a well-marked nucleus. Reissner's fibre passes through the optic ventricle, reënters the brain tissue, and again emerges into the fourth ventricle. Thus *Petromyzon* furnishes the connecting link between the condition in the Gnathostomes and *Amphioxus*. In the latter the largest and most anterior of the colossal dorsal cells lies across the central canal, is in direct connection with the pigment spot, and sends its axon caudad through the cord in the median plane just ventral to the canal. This axon probably represents Reissner's fibre.

There are many lines of evidence which lead me to assign the function I have to this apparatus.

1. The cells are in direct connection with the endings of the optic nerve, and with the cerebellum. The axons pass by the shortest path posteriorly through the central canal, and probably out through the ventral roots to the musculature.

2. When the fibre is cut in Sharks or Dogfish they evidence an inability to respond quickly to optical stimuli.

3. In the vertebrates of the cave fauna the apparatus degenerates as the eye degenerates.

4. In no animal does the apparatus reach complete development until just before the animal attains free life.

5. In those animals which are sluggish at hatching (*Petromyzon*, *Amia*), the apparatus is not fully developed until a considerably later period.

6. In those mammals which are born blind (Mouse, Kitten), the apparatus is not fully established until about the time the eyes become functional.

7. In any one group, as the Teleosts, the apparatus has its highest development in those species which are most active.

8. The corpora quadrigemina of higher vertebrates are concerned only with reflex functions; therefore this apparatus must have a reflex function.

Such a short circuit avoiding the loss of time in passing through a chain of neurones must be of great importance in saving time, amounting perhaps to a considerable fraction of a second. An animal suddenly presented with some optical evidence of danger from which it recoils in fear, does so reflexly, calling into use this apparatus. When we consider that in the struggle for existence the saving of a fraction of a second is often a matter of life or death, it becomes evident that this apparatus has played an important part in the survival of the fittest, and in the whole evolutionary process throughout the vertebrate series.

XII. THE SIGNIFICANCE OF THE SYNAPSIS STAGE OF THE GERM CELLS.

THOS. H. MONTGOMERY, JR.

IN the germinal cycle of the *Metazoa* may be distinguished in succession the following main stages: the conjugation of the maternal and paternal cells (fertilization), a number of generations of ovogonia (or spermatogonia), then the growth period, and finally the stage of the two maturation divisions. The reduction in the number of the chromosomes, *i.e.*, the formation of bivalent chromosomes, is not effected by either of the maturation mitoses, but during that portion of the growth period known as the synapsis stage. The bivalent chromosomes are formed by a union, end to end, of every two univalent chromosomes, as I have shown in a paper on the spermatogenesis of *Peripatus*, just published, and in another on the spermatogenesis of the *Hemiptera*, now in press.

Heretofore no one has shown exactly how the bivalent chromosomes are produced, and no one has given any adequate

explanation for the reason of their formation. My comparative studies on the spermatogenesis of a considerable number of species of *Hemiptera*, which have brought to light certain facts of importance for determining these questions, render it probable that in the process of formation of the bivalent chromosomes we have a conjugation of paternal with maternal chromosomes. This would then be the final stage in the fertilization of the germ cells ; it would be a conjugation of the chromosomes of different parentage producing a rejuvenation of them as metabolic centers of the cell ; and this rejuvenation finds its expression in the great changes of the growth period. Then, probably, the reduction division takes place, in order to separate again the conjugating chromosomes, as two conjugating *Infusoria* unite and then separate after the accomplishment of the rejuvenescence.

In the space of a short abstract it is not possible to give the evidence for these conclusions.

XIII. A STUDY OF THE PHENOMENA OF FERTILIZATION AND CLEAVAGE IN ETHERIZED EGGS.

EDMUND B. WILSON.

A.

IF fertilized eggs of *Toxopneustes*, after the formation of the cleavage-figure, be placed in a 2.5% solution of ether in sea water, the astral rays quickly fade out, as was long since observed by O. and R. Hertwig in sea-urchin eggs treated by solutions of chloral hydrate or sulphate of quinine. The clear hyaloplasm masses forming the astral centers are thus left as well-defined, slightly irregular, non-radiate areas, connected by the spindle-area. If the eggs are replaced in sea water, the rays are rapidly redeveloped and cleavage may proceed nearly or quite normally. Even if left in the ether solution, however, the nuclear division may be completed, the daughter-nuclei being re-formed and growing to their normal size, *but no cytoplasmic division occurs*, — a result similar to the earlier

ones of Demoor on the division of plant cells *in vacuo*, or in an atmosphere of CO_2 , and to those of Loeb and Norman on *Arbacia* eggs in sea water concentrated by the addition of a small percentage of chloride of sodium or magnesium. If the strength of the ether solution be now somewhat reduced, by evaporation or by the addition of sea water, the asters reappear, though not attaining full development, and progressive nuclear division may occur without cytoplasmic cleavage. In this case the egg may give rise to a syncytium, containing from four to sixty-four or more nuclei, which migrate towards the periphery so as to take up nearly the same position as they would have had in a segmented blastula. This phenomenon strikingly recalls that which normally occurs in the cleavage of many arthropod and some coelenterate eggs. At each nuclear division an attempt is made at a corresponding cytoplasmic division, but this is usually unsuccessful; or, in case division occurs, the cells subsequently fuse together to repeat the attempt at the next nuclear division. This, again, is closely similar to the ineffective early attempts at cleavage in such eggs as those of *Renilla*. If the eggs be replaced in sea water when the process is not too far advanced (4-32 nuclei), cleavage may occur of a multiple type almost exactly like that occurring in *Renilla*, and a normal blastula may arise; but the cleavage is often irregular or incomplete.

These observations support the conclusion indicated in the preceding paper, that the astral rays are not fixed and permanent structures, but an expression of a form of cytoplasmic activity, partly in the nature of protoplasmic currents, that may be inhibited by temporary paralysis of the cytoplasm. They indicate also that the astral rays are connected with cytoplasmic rather than with nuclear division, and support the interpretation, offered by the author many years ago, of the variations of cleavage observed in *Renilla*.

B.

If *Toxopneustes* eggs be placed in 2.5% ether solution one minute after fertilization, formation of the sperm-aster is completely suppressed. The sperm nucleus, however, slowly moves

inwards and gradually enlarges, becoming finally (1-2 hours) as large as the egg nucleus and indistinguishable from it. In some cases, though this is not very common, the two nuclei approach and finally completely fuse to form a typical cleavage nucleus. If in the earlier stages of the process (before union of the germ nuclei and while the sperm nucleus is still not more than two-thirds the diameter of the egg nucleus) the eggs be replaced in pure sea water, the sperm-aster is rapidly developed, centering in a point at one side of the sperm nucleus, and development may proceed normally; but this result was never obtained after the germ nuclei had united, probably because the action of the ether had been too prolonged. In a few cases, after replacing the eggs in sea water, the sperm-aster was observed to divide and form an amphiaster before union of the germ nuclei. In this case the sperm nucleus at the time of union had assumed the vesicular form, though still somewhat smaller than the egg nucleus. One such case was followed out and found to give rise to a normal larva. In such cases the effect of the ether has been to transform the type of fertilization from that characteristic of the sea urchins into that observed in starfishes, or in many worms and mollusks, where an amphiaster is formed before union of the germ nuclei and the latter are approximately equal at the time of union.

The foregoing facts show, in general accordance with the early work of O. and R. Hertwig, that growth of the sperm nucleus and approach and fusion of the germ nuclei may take place quite independently of the sperm-aster; further, that approach of the nuclei is probably not a simple chemotactic phenomenon, since it is very greatly delayed by etherization of the egg.

C.

In some of the etherized eggs, after replacement in sea water, the nucleus failed to divide at the first cleavage, the whole of the chromatin passing to one pole and re-forming as a single nucleus. Such eggs divide into a nucleated and a non-nucleated half, the latter containing only an aster, as in the case of some of the non-nucleated egg fragments fertilized

by a single spermatozoön observed by Boveri. In such cases the asters in both halves multiply progressively at the same rate, but complete division occurs in only the nucleated half. In the non-nucleated half, however, each aster becomes surrounded by a deep constriction which afterwards fades out. This result stands intermediate between those of Boveri and Ziegler. As in the case of the magnesium eggs, an aster unaccompanied by nuclear material forms a division center of the surrounding cytoplasmic area, but is here apparently unable to effect complete division in the absence of chromosomes.

XIV. EXPERIMENTS UPON THE INFLUENCE OF THE SEXUAL CELLS UPON THE SOMATIC CELLS.

GEORGE WILTON FIELD.

1. Of the sexual cells of one individual upon the somatic cells of another individual.

This is generally held to be a phase of Telegony. Several instances have been cited by Gadow, Nathusius, and Bulman to show that the spermatozoa affect the somatic cells which secrete the eggshell. Thus, the shell of eggs of fowls which normally lay brown eggs are said to become lighter in color when the birds are mated with a male of a breed which lays white eggs; and conversely, white eggs become brownish, if the birds are mated with a male of a breed which lays brown eggs.

Our experiments were carried on at the Rhode Island Agricultural Experiment Station upon Leghorn pullets (white eggs) mated to a Light Brahma cock, and upon Light Brahma pullets (brown eggs) mated to a Leghorn cock. By means of trap nests the series of eggs laid by each individual was followed; no departure from the ordinary normal color of the egg-shell could be observed.

2. Of the sexual cells upon the somatic cells of the same individual.

In addition to observations upon the changes in the secondary sexual characteristics induced by castration, the effect of absorption of testicular material through the peritoneum was the end in view.

Upon castration the testes of the cockerel were returned free into the abdominal cavity. After six weeks a living ripe testis, one inch long, was introduced into the abdominal cavity through an incision anterior to the last pair of ribs. The experiment was conducted on a scale too small to absolutely determine how far the results were due to direct regeneration of the testes, and how far to absorption of the introduced testicular material. Two points, however, were demonstrated: (1) that the testes do regenerate; (2) that the introduced testicular material was absorbed. In one case it seemed clear that the secondary sexual characteristics developed solely as the effect of the absorption of the introduced testicular material, without regeneration of the testes. In another individual similar conditions were strongly indicated, but were obscured by a pathological growth.

XV. THE MORPHOLOGICAL PHENOMENA INVOLVED IN THE CHEMICAL PRODUCTION OF PARTHENOGENESIS IN SEA URCHINS.

EDMUND B. WILSON.

In accordance with Loeb's important discoveries on *Arbacia*, unfertilized eggs of *Toxopneustes*, when treated by the magnesium chloride method, may segment and give rise to free-swimming blastulas, gastrulas, and Plutei. There is always a considerable proportion of abortive and monstrous forms, and none of the stages are exactly like those arising from fertilized eggs, though often closely similar to them. The Plutei possess, however, the characteristic arms, pigment, skeleton, and divisions of the gut. That these eggs have not been accidentally fertilized is proved by the fact, demonstrated to the society by an exhibition of sections, that during cleavage they

show but half the usual number of chromosomes, namely, eighteen instead of thirty-six. The same conclusion is reached by the study of the other internal phenomena, which differ in a characteristic way from those occurring in normal fertilization, though showing an interesting parallel to them.

The eggs, even of the same individual, show a very high degree of variability in their response to the solution. Great numbers of incomplete or abnormal forms of mitosis occur. The most interesting of these are cases in which the nucleus becomes the center of formation of a single aster (monaster), which never resolves itself into an amphiaster but nevertheless passes through periodic changes parallel to those occurring in complete mitosis. Thus, such an aster may appear, nearly disappear, and reappear as many as six times in succession, the nucleus simultaneously disappearing and re-forming. In such cases the chromosomes divide, probably at each disappearance of the nucleus, and may thus become very numerous, without division of the nucleus or of the cell body. In other forms of incomplete mitosis the single aster may give rise to an amphiaster, but the nucleus fails to divide.

In all the eggs capable of development, the initial change is an irregularity in the cytoplasmic meshwork, followed by the appearance of a *primary radiation* centering in the nucleus, the gradual formation of a perinuclear clear zone of hyaloplasm, and the growth of the nucleus. In many of the eggs a number of separate asters (cytasters, equivalent to the "artificial astrospheres" of Morgan), having no direct relation to the nucleus, are formed in the cytoplasm in addition to the primary radiation. At the centers of these asters hyaloplasm likewise accumulates. Growth of the nucleus is followed by disappearance of the nuclear membrane, the rays of both the primary radiation and of the cytasters meanwhile becoming much reduced and in some cases nearly disappearing. After a short pause this is followed by a redevelopment of the rays, and in typical cases the nuclear area (the center of the former primary radiation) has now formed two centers of radiation, producing a typical amphiaster. When no cytasters are present (a relatively rare case) cleavage may proceed nearly as in

normally fertilized eggs, but in many cases complete cytoplasmic division does not occur until after two or more nuclear divisions. Thus arise some of the forms of multiple cleavage observed by Morgan and Loeb. If cytasters be present, one or more of them may participate in the nuclear division, thus forming triasters, tetrasters, etc.; but such eggs are probably incapable of producing an embryo. When the cytasters do not establish a connection with the chromosomes, they nevertheless form, in many cases, ineffective centers of cytoplasmic division, *i.e.*, cleavage furrows appear between them but afterwards fade out. Apparently strong evidence was, however, obtained that in some cases complete division may occur around asters unconnected with nuclear material. In any case the cytasters persist for some time and may progressively multiply by division. The first division actually observed takes place nearly synchronously with that of the cleavage asters, at a time when the daughter-nuclei have been formed and are rapidly enlarging. Division of the asters is in both cases preceded by a great reduction of the astral rays, leaving the clear hyaloplasmic central mass surrounded by only short irregular rays, and at the same time the aster migrates out towards the periphery of the egg. The mass then draws apart into two, and recrudescence of the rays from the two centers ensues. A discrepancy, not yet fully cleared up, lies in the fact that, although the cytasters divide synchronously with the cleavage asters, they have not been observed in the living eggs to divide at the time the dicentric nuclear figure is first formed; but the study of sections indicates that this is probably owing to a gap in the observations. The cytasters ultimately disappear.

Asters are formed also in *enucleated fragments*, obtained by shaking the unfertilized eggs into pieces, and such asters may also progressively divide, though no case was observed of cleavage, or even an attempt at cleavage, in such fragments. Sections show that all the asters, whether cytasters or nuclear asters, or those formed in enucleated fragments, contain centrosomes which have the typical staining capacity and granular structure observed in normally fertilized eggs. In the cytasters, however, they are usually smaller than in the nuclear

asters, and those in the enucleated fragments are smaller still and often not demonstrable.

The observations indicate that the astral rays, whatever be their other functions, are in part an expression of centripetal currents of hyaloplasm (continuous or interalveolar substance), which lead to the formation of the perinuclear hyaloplasmic zone, and of the clear centers of the cytasters — a conclusion essentially in agreement with the early views of Fol. They show further that the asters (centrosomes) must be regarded as centers of cytoplasmic division, though not ordinarily effective unless connected with nuclear material. They seem to leave no doubt, finally, of the formation *de novo* of functional asters and centrosomes, capable of division, and show that such formation may be entirely independent of the nucleus.

XVI. METAMORPHOSIS IN THE HERMIT-CRAB.

M. T. THOMPSON.

In *Eupagurus longicarpus* only the first six larval stages are distinct: the four zoëas, the important glaucothoë, and the first of the adolescent stages. In the zoëas and the early part of the glaucothoë stage, the "livers" or midgut diverticula are cephalic and thoracic. There are two pairs of these; a pair of *Lesser Lobes* opening dorsally into the stomach, and a pair of somewhat four-lobed *Greater Lobes* opening laterally into the stomach.

During the glaucothoë stage, however, three of the divisions of the greater lobes become atrophied. The fourth or posterior division, at about the time of the second or third day in the shell, grows back into the abdomen. But the lobe of the right side of the body crosses under the intestine to the left, so that both lobes lie on the left of the intestine, which is thrown to the right. At this time the bladders of the Green Glands also migrate into the abdomen. Then the appendages which will be lost become atrophied, and the body musculature alters to the adult type. So the glaucothoë, which was

at first Macruran, attains the Eupagurid structural plan before the moult to the sixth stage occurs.

The duration of the glaucothoë stage is dependent on the time of entering the shell. Specimens which take the shell within the first twenty-four hours after the moult from the fourth zoëa, spend only *four* or more often *five* days in the stage. A delay of four days before the shell is taken prolongs this period to *six* or, in a few cases, to *seven* days. In fifty glaucothoë kept without any shells, some remained in the stage the minimum *four* days, but the majority remained *six* and *seven* days, and one remained *eight* days.

The sixth and following stages introduce no important changes in structure, except the branching of the liver. In this branching, however, the majority of the diverticula of the right lobe go to the right under the intestine, so that this lobe, *in the adult*, apparently lies on this, its own side, of the intestine. The lesser lobes branch later and finally form a small tuft of tubules on each side of the stomach.

XVII. ESSENTIAL FACTORS IN THE REGENERATION OF PLANARIA MACULATA.

CHARLES RUSSELL BARDEEN.

REGENERATION of a new whole individual from a small piece of a parent individual depends in *Planaria maculata* upon the presence in the piece of a part of the central nervous system and a part of the intestinal system. A small piece containing these parts will regenerate from them new typical intestinal and nervous systems. At the same time the parenchyma in the vicinity of the cut surface becomes increased in amount, and is symmetrically differentiated in relation to the new intestinal system. A head may thus be formed anterior to a new axial gut, and lateral and tail areas may be restored. Polarity of the piece is determined by the central nervous apparatus which it contains.

A new pharynx is formed just posterior to the point where intestinal contents collect when the whole piece contracts.

The pharynx may be formed in a region of the piece at some distance from the cut surface. Head, lateral, and tail areas are differentiated only at a cut surface.

The reproductive organs are not regenerated. Instead, they disappear from a small piece isolated from a sexually mature worm. The tail cut from a planarian in which the reproductive organs are developing will give rise to regenerative forces which overpower the forces giving rise to the sexual organs. Regeneration is equally rapid in sexually mature and in sexually immature worms.

In regeneration in this animal the tissues seem to be specific, except that the new musculature probably comes from parenchyma cells.

A full account of the Physiology of Regeneration in these animals is given in the *American Journal of Physiology*, Vol. V (1901), p. 1.

XVIII. THE HISTOGENESIS OF THE PERIPHERAL NERVOUS SYSTEM IN *SALMO SALAR*.¹

ROSS GRANVILLE HARRISON.

CELLS provided with pseudopodia-like processes wander out singly from the dorsal surface of the medullary cord, and collect together between the myotomes and the cord into small groups, the spinal ganglia. Here the cells remain for some time undifferentiated, but are transformed later into bipolar cells, of which the centripetal processes grow into the side of the medullary cord to form the dorsal roots.

Neuroblasts may be distinguished at an early stage as round or polyhedral cells, lying in the outer zone of the cord. At this period the cord is made up chiefly of epithelial cells, the forerunner of the ependyma. These cells are still undifferentiated, no specialized "*Randschleier*" being present. As the axones grow out from the neuroblasts, they bore their way

¹A full account of this work is published in the *Archiv für mikroskopische Anatomie*, January, 1901.

through the substance of the epithelial cells, which with the continued growth of new fibres become more and more honey-combed. Their outer zone is finally transformed into a fibrous framework, the "*Randschleier*," which accordingly owes its structure to the activity of the growing nerve fibres, and is not pre-formed.

The dorsal cells or giant cells of Rohon arise in the dorso-lateral portion of the cord next the outer limiting membrane. They elongate, and for the most part each cell gives rise to an ascending and a descending nerve fibre forming the beginning of the dorsal funiculi. Gradually the cells leave their original position and wander to the dorsal mid-line of the cord. Through this movement the cells become unipolar, remaining connected with their fibres by a slender process, which divides in T-fashion at the point where the longitudinal fibres begin. A large number of the cells form peripheral nerves also, which are segmentally arranged. The dorsal cells are homologous with the "*Hinterzellen*" found in *Petromyzon*, and with the bipolar cells of medium size in the cord of *Amphioxus*. They are to be regarded as a primitive type of sensory cell identical in function with the spinal ganglion cells, with which they are genetically related.

XIX. THE SPERMATIC AND MESENTERIC ARTERIES OF DIDELPHYS VIRGINIANA (KERR, LINN.).

C. F. W. McCLURE.

In mammals other than marsupials the anterior mesenteric artery supplies the small intestine and the proximal end of the large intestine. The posterior mesenteric artery is given off from the posterior division of the abdominal aorta, and supplies the large intestine. In a large number of mammals the internal spermatic arteries are given off from the aorta about midway between the renal and posterior mesenteric arteries.

In *Didelphys* and other marsupials, so far as known to the writer, the anterior mesenteric artery supplies both the small and

large intestines. In *Didelphys* and other marsupials the posterior mesenteric artery *is not present*. Also in *Didelphys* and other marsupials the internal spermatic arteries are given off from the posterior division of the aorta, and at a point which coincides with the point of origin of the posterior mesenteric artery in other mammals.

In an adult *Didelphys* killed during the breeding season the writer found present two pairs of functional internal spermatic arteries. The anterior pair was given off from the aorta about midway between the renal and posterior pair of internal spermatic arteries. The posterior pair, the so-called internal spermatic arteries of marsupials, was given off from the aorta in the usual manner, as mentioned above. More recently the writer has found another adult female *Didelphys*, in which, in addition to two pairs of internal spermatic arteries, a large *posterior mesenteric artery was present*.

In this individual the posterior mesenteric artery arose from the aorta as a single vessel, and at a point which coincided with the origin of this vessel in other mammals. On arising from the aorta the vessel passed ventrad through a foramen in the vena cava, and was distributed to the large intestine. The anterior mesenteric artery in this individual supplied the small intestine and the proximal portion of the large intestine.

The relations of the spermatic arteries were as follows:

The anterior pair of internal spermatic arteries arose from the aorta, and was distributed to the ovaries as in the above-mentioned case. These arteries appear to be the homologues of those spermatic arteries which in many other mammals arise from the aorta about midway between the renal and posterior mesenteric arteries. The posterior pair of internal spermatic arteries in this opossum were *branches of the posterior mesenteric artery*, and were given off from this vessel near its point of origin at the aorta.

It appears to the writer as though in the marsupials, as the result of an arrested development of the original internal spermatic and the posterior mesenteric arteries, a new collateral circulation has been established to the genital organs

and large intestine. The collateral circulation to the large intestine has apparently been established through the anterior mesenteric artery; that to the ovaries, through vessels which may have been formed as the result of a modification of the posterior mesenteric artery.

XX. SOME FACTS CONCERNING REGENERATION AND REGULATION IN RENILLA.

H. B. TORREY.

DURING the past summer experiments were carried on at Beaufort, North Carolina, preliminary to a more complete investigation of the processes of regeneration and regulation in *Renilla*. It was hoped that *Renilla*, being a polymorphic colonial form,—an *aggregate of polyps and zooids*,—would behave like a simple metazoan individual, and at the same time offer surer landmarks, during regulative processes, than a metazoan individual—an *aggregate of cells*; for changes in polyps as a whole may be perceived more clearly than changes in their component cells.

The results may be summarized as follows:

1. *Renilla* colonies may regenerate lost parts readily.
2. They exhibit a strong polarity. When a peduncle is removed by a transverse cut an axial polyp is never regenerated in its place, and *vice versa*.
3. There is an anterior limit beyond which anterior pieces do not regenerate posteriorly, and a posterior limit beyond which posterior pieces do not regenerate anteriorly. These correspond to the limits of the budding zone.
4. The colonies regulate themselves in a plastic fashion when cut in certain ways, obliquely, for instance. It is thus possible to obtain two new colonies, one of which retains the original peduncle with a lateral polyp displaced into the position formerly occupied by the axial polyp. Whether or not the colony develops symmetrically around this new axis is not known.

If the oblique cut makes with the colonial axis an angle larger than forty-five degrees, there is no displacement of the lateral polyp, the extirpated axial polyp regenerating as though it alone had been removed by a transverse cut.

5. When a lateral group of polyps is removed by a longitudinal cut, it regenerates a new peduncle approximately at a right angle to the cut surface, and approximately in the axis of the chief lateral polyp of the group. The future of such pieces is unknown. This is a case of heteromorphosis.

XXI. SOME POINTS IN THE BRAIN OF LOWER VERTEBRATES.

J. B. JOHNSTON.

THE central olfactory apparatus of *Petromyzon* presents, in all important features, an extraordinary resemblance to that of *Acipenser*. In *Petromyzon*, on account of the great buccal apparatus, there has occurred a sort of telescoping of the olfactory lobes and areas upon the striatum and thalamus as fixed points. The so-called cortex, described by Friedrich Mayer, is nothing else than the epistriatum.

The cells of the olfactory lobe present more primitive characters in *Petromyzon* than in *Acipenser*. The mitral cells are only slightly differentiated, while the stellate and other cells are very numerous and send their neurites, along with those of the mitral cells, to the olfactory nuclei of the fore-brain. Similar categories of cells have been described in Amphibia (P. R. Cajal) and reptiles (Edinger's "Lobuscortex"), although differently interpreted. The numerous, slightly differentiated cells in the olfactory lobe of *Petromyzon* and *Acipenser* represent the material from which the highly differentiated elements of the olfactory lobe of higher vertebrates have been developed.

Several authors have pointed out the close connection between the cerebellum and acusticum in fishes. The study of the minute structure shows that the cerebellum is derived

directly from the front end of the acusticum. Evidence for this from *Acipenser*:

- a.* Gross continuity of cerebellum and acusticum.
- b.* The root fibres of the fifth, eighth, and lateral line nerves enter and end in both.
- c.* The several categories of types of cells in the cerebellum — Purkinje cells, granules, and cells of the second type — are strictly homologous with similar cells found in the acusticum.
- d.* The development of the Purkinje cells in the acusticum from the typical large cells of that nucleus is in actual progress and may be studied in all its stages.

Additional evidence in *Petromyzon*:

- a.* The Purkinje cells in the cerebellum are not well developed, and their neurites run to the base of the mid-brain, possibly having the same destination as the internal arcuate fibres from the acusticum.
- b.* The *tractus tecto-cerebellaris* seems to be absent and the *tractus lobo-cerebellaris* is small.
- c.* The cerebellum is little more than a dorsal arch and commissure from the front end of the acusticum.

XXII. ASEXUAL REPRODUCTION OF PLANARIA MACULATA.

WINTERTON C. CURTIS.

THE fission of *Planaria maculata*, while it does not differ essentially from the type found in other planarians where fission occurs, is of just the right sort to complete a very interesting series and connect the fission of land planarians, which is hardly more than a fragmentation, with the fission of *Planaria fissipara*, in which the organs are completely formed before the new individuals separate. This series is as follows: (1) Land planarians, in which pieces of varying lengths are pinched off from the posterior end; (2) *Planaria maculata*, which divides always at the same place behind the pharynx, with no preformation of organs; (3) *Planaria subtentacula*

(Zacharias, *Zeit. f. Wiss. Zool.*, 1886, Bd. XLIII, pp. 271-275), where there is some rearrangement of the gut and the pharynx is partly developed before separation; (4) *Planaria fissipara* (Kennel, *Zool. Jahrb.*, Abth. f. Anat. u. Ontog., 1888, Bd. III, pp. 447-486), in which a complete worm is developed out of the posterior third of a large specimen and both reach normal proportions before separation. In the last three cases the division occurs at a corresponding place.

The division in *Planaria maculata* seems to be brought about by a contraction of the circular muscles, which pinches the individual in two a short distance behind the pharynx. The cut ends of either piece are as though they had been produced by a knife-cut, and examination of sections shows that the parenchyma at the scar is actually naked. There is nothing like a furrow on the outside previous to the division, which, nevertheless, is a regular and normal reproductive process and not induced by any ordinary irritation or mutilation of the animals. The large number of pieces in various stages of regeneration, found in collecting, is sufficient evidence of the occurrence of the fission under natural conditions.

Worms will not divide in the laboratory to any considerable extent unless well fed. If the water has become foul and is replaced by fresh, a considerable number of specimens will usually be found divided within the next twelve hours. The division usually occurs at night, whether the dishes are shaded or not. The morpholaxis of the head and tail pieces resulting from a division is rapid. Tail pieces may reach almost the normal proportions and re-divide in from five to six days if well fed, heads in not less than ten days. There is no regular interval between the divisions.

In certain localities this species does not possess reproductive organs at any time during the year, but has during the summer months an active period of asexual multiplication. In other localities the worms develop these organs in the fall and lay eggs in the spring; and although all the specimens are without these organs at the end of the summer, asexual reproduction has never been observed. In another sexual locality the worms, when they are without reproductive organs at the

end of the summer, do reproduce asexually to a considerable extent. Later this ceases and the reproductive organs develop. These statements are based upon observations extending over from two to three years.

A possible explanation is that the asexual reproduction may be substituted for the sexual in certain localities during considerable periods, but further data are necessary to confirm this.

XXIII. VARIATION AMONG HYDROMEDUSAE.

CHARLES W. HARGITT.

OBSERVATIONS upon variations among the Hydromedusae seem to have been of comparatively limited extent. References to the subject are to be found in the writings of Ehrenberg, Forbes, Agassiz, Hincks, Romanes, and later by Agassiz and Woodworth, but except in the last-named paper they are rather incidental and fragmentary.

Of my own observations only the barest abstract and summary can be undertaken in this connection.

Among the genera studied the principal have been as follows: *Pennaria*, *Eucope*, *Obelia*, *Margelis*, *Gonionemus*, *Nemopsis*, *Rhegmatodes*.

The principal organs examined were: (1) The Chymiferous Canals, (2) Tentacles, (3) Gonads, (4) Otocysts. Among these the greatest range of variation was noted in the tentacles, as might naturally be expected, in some cases reaching as high as 90 per cent. In the forking and doubling of tentacles there was least, rarely exceeding 5 per cent, and indeed seldom reaching that ratio; in *Gonionemus* 3 per cent.

In the looping, branching, and anastomosing of chymiferous canals there was great variation in different genera, in some being almost *nil*, while in others (*Eucope* and *Gonionemus*) varying from 5 to 10 per cent.

Considerable variation was found in the gonads, though less than in the other organs already noted, varying in different genera from 2 to 5 per cent. While considerable variation

was evident in the number, arrangement and correlation of the otocysts, no attempt has been made to ascertain the exact ratio, owing to the difficulty attending this determination in preserved specimens.

The following summary will express in a general way some of the more evident conclusions reached :

1. Variation among Hydromedusae is of wider extent than had been suspected.
2. It is much greater in some genera than in others.
3. It seems to be much less symmetrical and correlated than among Scyphomedusae.
4. Many phases of variation appear to be wholly devoid of correlative and adaptive aspects.

XXIV. EXPERIMENTS ON MODIFYING THE NORMAL PROPORTION OF THE SEXES IN THE DOMESTIC FOWL.

GEORGE WILTON FIELD.

THIS is a brief report on a series of experimental attempts to ascertain the factors which determine sex.

The normal proportion given by Darwin from observation of 1001 chicks during eight years was 94.7 males to every 100 females. From 2105 chicks during two years, we found the proportion to be 80.6 males to every 100 females.

These figures lead us to query whether the normal proportion may not have changed during the past forty years as a result of the breeders' desire to produce a larger proportion of females.

In the experiments attempts were made to isolate the factors so that the effects of each could be observed :

1. Absolute age of parents :

9 young females mated to male of same age.
9 old " " " " " " "
2. Relative age of parents :

9 old females mated to young male.
9 young " " " old "

The proportion practically coincided with the normal except in the case of young females mated to old male, where a slight increase in the number of males appears.

3. Malnutrition (a result of feeding to one-half usual amount): a very marked increase in number of males; rate of 176.6 males to every 100 females.

4. Scarcity of males, *i.e.*, polygamy: 20 females mated to 1 male, a marked increase in number of males; 139.6 males to every 100 females.

5. Conditions connected with time of year: 650 chicks hatched between March 15 and May 15 gave a smaller proportion of males, — 73.9 males to every 100 females; while 471 chicks hatched between May 15 and July 15 gave a larger proportion of males, — 88.4 males to every 100 females.

It is to be understood that these figures are for a relatively small number of cases; it is hoped to extend them to at least 10,000 individuals.

XXV. NOTES ON VARIATION IN THE SHELLS OF PURPURA LAPILLUS.

R. P. BIGELOW AND H. S. CONANT.

Purpura lapillus is a species that presents great variations in its diagnostic characters. It was thought, therefore, that a study of its variations by statistical methods might be of value in defining more exactly the limits of the species, and might also bring to light facts of general biological interest. Collections were made at Prince's Cove and on the mainland opposite Clark's Ledge, Eastport, at Kennebunk Beach, at Bass Rocks, Gloucester, and at Newport. The sexes were separated for each locality, and the following characters were measured:

(1) Angle of the apex and nuclear whorls, or nuclear angle; (2) angle of the apex and the last whorl, or adult angle; (3) total length; and (4) length of spire from the apex to the posterior margin of the opening of the shell. Record was made also of the presence of (5) imbrications, (6) sutures, (7) ribs, (8) teeth, (9) of the curvature of the columella, and (10) of the weight. Perhaps the most obvious variations are in the color and the thickness of the shell, but no satisfactory method was

found for measuring these quantities. A special instrument is being constructed which, it is hoped, will overcome the difficulty in regard to thickness.

A preliminary study of the shells from Eastport and Gloucester shows that for characters (1) to (4) the variations may be represented by curves that are approximately normal. The curves for the two stations at Eastport fit together pretty closely; while they differ distinctly from the curves for the Gloucester specimens, the difference of the means being greater than the standard deviation for each locality. In each case the female shells showed, on the average, a wider angle and a shorter spire expressed in per cent of total length, than the corresponding males, and the same is true for the Gloucester shells as a whole compared with the Eastport shells.

As a measure of variability the coefficient of variation $\left(cv. = 100 \frac{\sigma}{m} \right)$ gives contradictory results and appears not to be applicable to measurements expressed in degrees of a circle. Judging from the standard deviations, the shells from Gloucester are somewhat more variable than those from Eastport. The relative variability of the males and females differs for the different characters, and for the same characters in different localities. In general, the females appear to be slightly more variable than the males.

XXVI. VARIATION AND ELIMINATION IN PHILOSAMIA CYNTHIA.

HENRY E. CRAMPTON.

SOME of the results were presented of a statistical study in the case of a large Saturnid moth, *P. cynthia*, of the variability of eliminated and surviving pupae and imagines. From a lot of nearly 1100 cocoons, only 310 living pupae were obtained; 632 contained dead pupae; while the remainder were shriveled or otherwise abnormal larvae or pupae. The living pupae were compared with an equal number of dead pupae with reference

to certain body characters: length, length of bust (to fifth abdominal segment), width and depth of bust, frontal stature of bust (ratio of middle to length), and sagittal stature of bust (ratio of depth to length). In addition the length, width, and stature of a typical organ, the left antenna, were determined. From a tabulation of certain bases of comparison — mean, standard deviation, coefficient of variability — it appeared: (1) That eliminated male pupae were on the whole more variable than the surviving males, and that the surviving females were far less variable than the dead ones. (2) Only 180 of the 310 living pupae produced perfect moths. The perfect male moths were from pupae which were far less variable than the others. This condition was reversed in the case of the females, yet the surviving females, though more variable than the eliminated ones, were not as variable as the eliminated female pupae. (3) The males of all groups were more variable than the females.

XXVII. THE ORIGIN OF TENTACLES IN GONIONEMUS.

H. F. PERKINS.

SOME interesting data have been secured from the study of the origin of tentacles in *Gonionemus*, a common Woods Holl hydromedusa. Specimens $\frac{1}{2}$ mm. in diameter having from 8 to 16 tentacles are found in early summer, and in examining these it was seen that there existed a definite relative position of the tentacles and sense organs.

Two pairs of tentacles, the radial ones, are of equal size. The other tentacles and sense organs are regularly graded from large to small, so that it is possible to determine their order of origin.

Looking at the marginal ring from below, in a normal medusa, each newly formed tentacle is seen to lie next to a sense organ and to precede it, as the hands of a watch move. Fig. 1 shows a typical 8-tentacled medusa. Tentacles I and II are radial in position; III follows I in the direction of the

hands of a watch; IV follows II, and the two pairs of sense organs, 1 and 2, lie as if the cells that were to form them had been crowded along to the right by the newly formed tentacles.

In an older specimen (Fig. 2) the successive pairs of tentacles and sense organs have arisen in corresponding positions, as is indicated by the numbers on the diagram. The origin

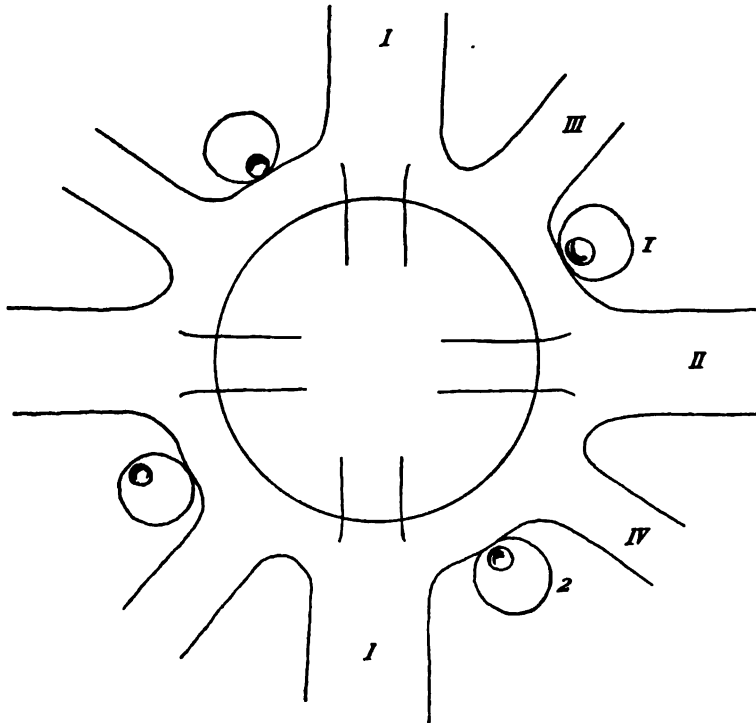


FIG. 1.

of tentacles and sense organs would seem then to be governed by an attempt at radial symmetry which is constantly interfered with by this sequence of formation from left to right, along the bell-margin.

In full-grown medusae there appears a striking conformity to this rule, with fewer exceptions than would be expected from the frequency of other variations in all parts of the creature.

The comparison of other and allied forms, with this rule in mind, may bring to light some interesting facts bearing on the correspondence of parts and on radial and bilateral symmetry.

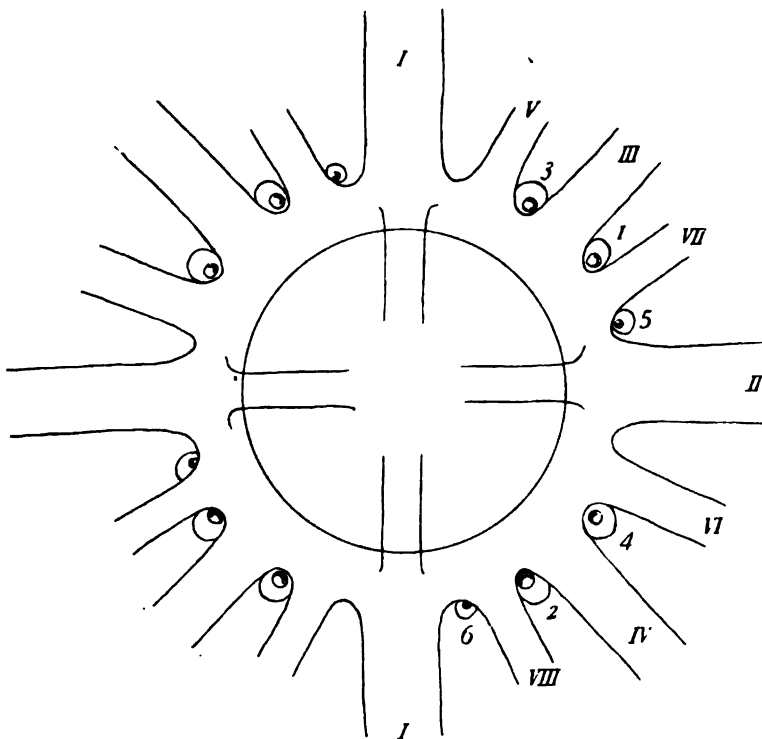


FIG. 2

